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Assessment of Nursery-Raised *Acropora cervicornis* Transplants in the Upper Florida Keys

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Nova Southeastern University
Department of Marine and Environmental Sciences

**Assessment of Nursery-Raised *Acropora cervicornis*
transplants in the Upper Florida Keys**

By
Matthew Ware

Submitted to the Faculty of
Nova Southeastern University Oceanographic Center
in partial fulfillment of the requirements for
the degree of Masters of Science with a specialty in:
Marine Biology and Marine Environmental Science

Nova Southeastern University

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Matthew Ware

Submitted in Partial Fulfillment of the Requirements for the Degree of

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ABSTRACT

Over the last 40 years, the Caribbean has lost half of its live coral cover, mostly in the form of *Acropora cervicornis* and *A. palmata*, due to disease, bleaching from rising water temperatures, and other stressors. To help restore these corals to reefs in Florida, the Coral Restoration Foundation (CRF) created nearshore nurseries and transplanted over 30,000 acroporid colonies across the Florida Keys. The objective of this thesis was to evaluate the growth, survivorship, and condition of nursery-raised *A. cervicornis* colonies that were part of two transplant projects: 1) photographic analyses of 17 past CRF transplant projects over the last seven years; and 2) a transplant experiment at Little Conch Reef to additionally assess the effects of depth, colony density, and the genetic composition of transplants. The photographic analyses included 2,428 individual colonies, 38 genotypes, and six reefs from 2007 to 2013. Results from the photographs were combined with one *in situ* monitoring effort that used SCUBA in 2014. In the Little Conch Reef experiment, 1,288 colonies from 14 genotypes were transplanted in October and November, 2013 at two depths (5m and 12m) in either cluster or thicket configurations. At each depth, clusters comprised 14 colonies, each placed within in 1m diameter radius, with ten monogenetic and six multigenetic structures. Thickets were 3.5m by 1.5m in size, with 10 colonies from each genotype forming its own subunit within the larger configuration. In June 2014, 963 additional colonies were added to the shallow site by stacking them on top of six existing clusters and one thicket to evaluate whether larger three-dimensional structures affected growth or survival. The Little Conch Reef experiment was monitored through January 2015. Results from the photographic analyses were: 1) maximum size of *A. cervicornis* transplants was approximately 40cm in diameter; 2) mortality increased after approximately two years; 3) despite high mortality, some colonies survived the duration of each project; and 4) frequent and long-term monitoring is required to assess factors that affect survival and condition. Results from the Little Conch Reef experiment suggest: 1) maximum skeletal diameter was unaffected by any of the treatments; 2) percent survival and percent live tissue were higher at the shallow site compared to the deep site, and similarly, the clusters outperformed the thickets, and multigenetic clusters outperformed their monogenetic counterparts; 3) location within the shallow site had an impact on survival and condition, with clusters

doing better on the south side than on the north; and 4) stacking did not positively impact growth, survival, or condition. In general, the sizes and condition of natural populations of *A. cervicornis* throughout the Florida Keys are similar to results from both experiments and with other transplant projects conducted in the Caribbean. Remarkably, despite high mortality in nearly all of the projects, small numbers of colonies transplanted for most projects, a few colonies survived to 2014/2015. These colonies have the potential to act as a “seed population” that might produce sexually dispersed larvae better adapted at surviving mortality events and asexual fragments that may be better acclimated to the stressors related to their location. Evidence of persistence in this species and expansion northward in Florida suggest that it is too early to consider coral reefs a lost cause, and that coral restoration holds promise for enhancing recovery of *A. cervicornis*.

Keywords: *Acropora cervicornis*, photographic analysis, coral reef restoration, transplanting technique, Coral Restoration Foundation, Florida Keys National Marine Sanctuary

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INTRODUCTION AND OBJECTIVES

Coral reefs are the most biodiverse systems on the planet with more phyla than any other ecosystem (Wilkinson 2008). They provide many valuable services that benefit human populations. Globally, they can be worth up to \$375 billion per year in coastal protection, fisheries, and tourism (Edwards and Gomez 2007). In terms of biological value, coral reefs hold 25% of the world's fish species (Hughes et al. 2010, Camargo et al. 2009). Over 3.6 million tourists per year support the Florida Keys economy, largely through use of coral reefs (Jackson et al. 2014). The entire Florida Reef Tract generates over \$6 billion (Johns et al. 2001). Their three-dimensional structure provides hiding and foraging space for everything from coral recruits to sharks. In addition, they are one of the few systems capable of constructing their own substrate, and in doing so, alter the flow of water around them (Sheppard et al. 2009).

Despite their local and global importance, coral reefs are in decline. In the last 40 years, the Caribbean has lost half of its live coral cover (Gardner et al. 2003, Jackson et al. 2014). Regional declines in coral cover were caused by disease (Aronson and Precht 2001), bleaching due to global warming (Eakin et al. 2010, Barton and Casey 2005), nutrient pollution (D'Angelo and Wiedenmann 2014), and coastal development (Wilkinson 2008, Hughes et al. 2010). Physical damage from storms and vessel groundings has impacted reefs at the local level (NMFS 2012, Alvarez-Filip et al. 2009). These stressors do not act independently. For example, increasing water temperatures weaken coral immune defenses through metabolic stress and may increase disease virulence (Harvell et al. 2002, Lesser et al. 2007).

The dominant reef-builders in Florida and the Caribbean, *Acropora cervicornis* (staghorn coral) and *Acropora palmata* (elkhorn coral), were prevalent throughout the Florida Keys until the late 1970s. An outbreak of White Band Disease in the 1970s and several coral bleaching events have reduced populations by as much as 95% throughout the Caribbean (Bruckner 2002, Aronson and Precht 2001). At Looe Key, Miller et al. (2002) estimated that 98% of the staghorn disappeared from 1983 to 2000. As a result, both species received Threatened status under the U.S. Endangered Species Act on May 9, 2006 (NMFS 2006).

There is currently much debate over the listing of *Acropora cervicornis* under the ESA because of its existing wide distribution and abundance both within and outside of U.S. jurisdictional waters (Shinn 2004, Precht et al. 2004, Bruckner and Hourigan 2000). Millions of colonies remain in Florida (Miller et al. 2008), though the colonies are relatively small (10-20 cm diameter) and scattered. Despite their persistence, it is clear that recovery to 1970s abundances has not occurred (Randall and van Woesik 2015, Vargas-Angel et al. 2003). Conditions that might promote natural recovery remain problematic. However, the development of nursery and transplanting techniques suitable for restoration of *A. cervicornis* provide optimism for recovery (NMFS 2014, Shinn et al. 2003, Rogers and Muller 2012, Young et al. 2012).



Figure 1. Map of *Acropora cervicornis* and *A. palmata* regional distribution. General distribution is shown in pink highlight (Acropora BRT 2005).

Restoration of *Acropora cervicornis* on a regional scale represents a huge challenge (Precht et al. 2005, Graham et al. 2014), but some successes have occurred locally (Johnson et al. 2011, Garrison and Ward 2012, Bruckner and Bruckner 2006). Fragments salvaged from storm damage or nursery-raised colonies were transplanted to

small patches of reef where they survived, but long-term monitoring has been limited. Transplanting storm-generated “corals of opportunity” remains controversial (Precht et al. 2005, Edwards and Clark 1999). A major benefit of coral nurseries is that they produce large numbers of colonies of a relatively large size (>10 cm with numerous branches) which can be used to restore sites damaged by ship groundings (Johnson et al. 2011), supplement natural recruitment, or to mitigate previous declines without overharvesting fragments from nearby natural populations (Monty et al. 2006, Garrison and Ward 2008, Lirman et al. 2010). Much of the technology used to develop nurseries and to transplant corals throughout southeast Florida was developed by, or in collaboration with, the Key Largo-based Coral Restoration Foundation (CRF) (Nedimyer et al. 2011, Young et al. 2012, Johnson et al. 2011). Since 2003, CRF has transplanted over 30,000 acroporid colonies on 14 reefs throughout the Florida Keys (Figure 2).

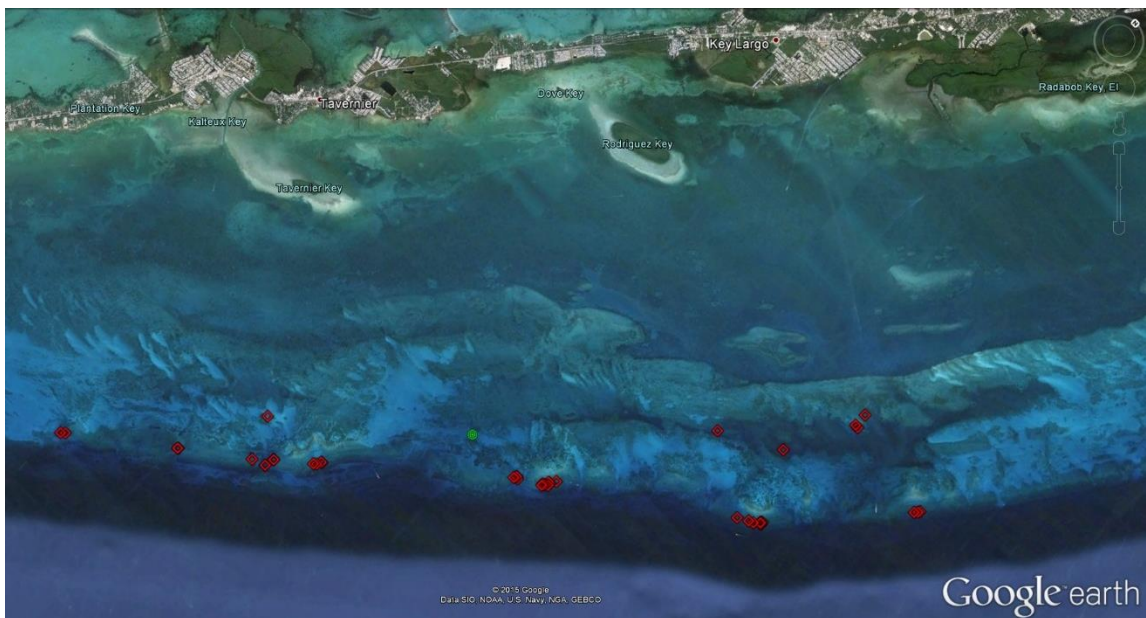


Figure 2. CRF staghorn coral transplant sites from Dry Rocks (right) down to Davis Reef (left) from 2003 to 2013 shown as red diamonds. Tavernier nursery is shown as the green circle.

Early Coral Restoration Foundation (CRF) transplanting projects were limited to fewer than 25 colonies due to permit restrictions. However, as the CRF nursery and transplanting techniques evolved to become more successful, a greater number of colonies across more locations were permitted. Currently, CRF maintains three nurseries

in the Keys and several more throughout the Caribbean. The foundation's approach is to transport and attach large numbers of corals from their nurseries to nearby reefs, followed by periodic maintenance that includes reattaching dislodged colonies or broken fragments, removing predatory snails, and photographic documentation.

An objective of this thesis was to assess the growth and survival of transplant projects conducted by the Coral Restoration Foundation over a period of seven years. In general, monitoring is not routinely conducted to assess the success of transplanted colonies or different transplanting methodologies (Precht and Robbart 2006, Wapnick and McCarthy 2006, Challenger 2006). Initially, numerous approaches were tested to determine the feasibility of restoration (Nedimyer et al. 2011, Miller et al. 2010). Questions included the scale and cost-effectiveness of coral nurseries, along with the attachment, growth, and whether or not reproduction of transplanted colonies occurred. Early monitoring efforts were done casually and often included general observations or photographs to meet permit compliance requirements. To help answer questions about the success of nursery-raised colonies transplanted onto offshore reefs, formal scientific monitoring programs are needed (Wapnick and McCarthy 2006, Edwards and Gomez 2007).

This thesis has two main components. First, a subset of past Coral Restoration Foundation projects in the Upper Florida Keys were assessed using historical photographs and present-day *in situ* resampling with SCUBA to evaluate the number, sizes, and condition of transplanted colonies. This work provided a multi-year assessment of nursery-raised *Acropora cervicornis* transplants, related to growth, survivorship, and condition. Variables include different coral genotypes, locations, and habitats. Second, a transplant experiment was conducted at Little Conch Reef starting in 2013 to determine the effects of depth, colony density, and genetic composition on growth, survivorship, and condition of *A. cervicornis*. This thesis reports on the initial year of monitoring results of the Little Conch Reef experiment.

METHODS

STUDY SPECIES

Staghorn coral, *Acropora cervicornis*, is one of two species in the genus *Acropora* that are found in the tropical Western Atlantic. Healthy colonies form highly branched structures up to 1 m across (NMFS 2014). Zooxanthellae give the colony a yellowish to brown appearance and are critical for nourishment (Porter 1976). Rapid growth rates (up to 12 cm yr⁻¹) exceed the reproductive rates of the endosymbiotic algae, causing the extending branch tips to appear white (Tunncliffe 1981, Shinn 1966).

Acropora cervicornis is cosmopolitan in the Caribbean, and is found from Venezuela to Florida and Mexico to Barbados. It prefers shallow fore-reef and patch reef environments down to 20 m depth, but has been recorded as deep as 60 m (Goreau 1959, Neigel and Avise 1983, NMFS 2014). Wave action at shallow depths supports asexual fragmentation as the dominant form of reproduction (Tunncliffe 1981, Highsmith 1982). Before their decline in the 1970s and 1980s, this mode of reproduction generated dense thickets (Gilmore and Hall 1976, Gladfelter et al. 1978). Today, colonies are typically found individually in low densities (Miller et al. 2008). *A. cervicornis* reproduces sexually via broadcast spawning only during a few nights from July to September, and can hybridize with its congener *A. palmata* (elkhorn coral) to produce *A. prolifera* (fused staghorn coral) (Vargas-Ángel and Thomas 2002, Fogarty et al. 2012). *A. palmata* populations in the Florida Keys are genetically isolated from other populations throughout the Caribbean (Baums et al. 2005). Evidence from Vollmer and Palumbi (2006) suggests *A. cervicornis* may be similarly regionally restricted. However, within Florida, *A. cervicornis* is genetically well-connected (Hemond and Vollmer 2010).

The *A. cervicornis* colonies utilized as in this study were raised in the Coral Restoration Foundation nursery located 5km off of Tavernier, Florida. The nursery was established in 2001 at a depth of 9m over a sandy bottom, in-shore of the outer reef tract. Fragments were attached to either fixed or suspended structures and permitted to grow to 10cm maximum diameter before being transplanted to the reef by CRF staff or CRF-trained volunteers. Among the nursery structures utilized to grow corals were disks, lines,

and trees (Johnson et al. 2011, Nedimyer et al. 2011). Upon transplant, colonies were secured to the reef with epoxy in a variety of configurations (such as Figure 3).

HISTORICAL PHOTOGRAPHIC ANALYSES

Photographic records of transplant projects from 2007 to 2013 were obtained from the Coral Restoration Foundation to develop a multi-year dataset for coral transplant growth, condition, and survivorship. The projects utilized different genotypes, reefs locations, and habitats. CRF conducted previous testing to identify genotype and maintains a record of lineage for fragments contained within their nurseries. Early projects (2007 – 2010) positioned colonies in a triangle approximately 1m across with a different genotype at each point (Figure 3A). Later projects placed monogenetic colonies around the perimeter of a circle approximately 1m in diameter (Figure 3B), or scattered individuals across the transplant site on suitable available substrate.

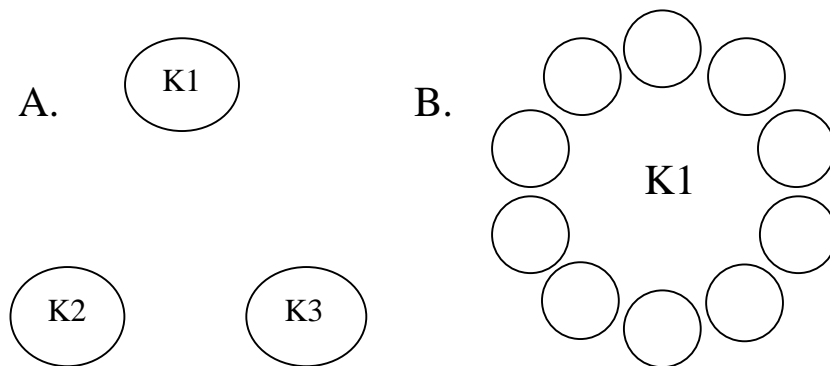


Figure 3. Transplanting configurations. Each circle represents a single colony. Text indicates genotype. “A” represents the triangular configuration, while “B” is the circular configuration. These configurations were repeated depending on the number of available or permitted fragments for the given project. The “scattered” configuration is not displayed.

Monitoring by CRF consisted of periodic visits to record general observations, attach broken fragments, and take photographs. No *in situ* growth or condition information was recorded. The photographs were matched to written project records to obtain the date of transplant and location, then screened for image quality, genotype tag identification, and scale information before being given a unique identifier code. Only projects with complete photographic records were utilized for analysis. To be considered “complete”, a project would have to have photographic documentation of all of the corals

transplanted, with scale information, from at least two different maintenance visits. 2,874 photographs from 17 projects were included in the analysis while 4,073 photographs representing 42 projects were excluded. The 17 included projects cover 2,428 individual colonies transplanted across six reefs from 38 genotypes and are representative of the body of CRF's work (Table 1). Different habitats where corals were transplanted included high-relief spur-and-groove, hard-bottom pavement, or patch reef topography (Florida Fish and Wildlife Conservation Commission 2014). Colony maximum skeletal diameter, perpendicular skeletal diameter, skeletal height, and percent living tissue, consistent with Miller et al. 2011, were obtained from the photographs using CANVAS software (ACD Systems, Inc. 2005). Genotype tags of known sizes or PVC scale bars were utilized for size reference (Figure 4). With known dimensions of the genotype tags or PVC scale bars, the skeletal dimensions of the colony could be extrapolated using a ratio of the colony dimensions in the image to the scale dimensions. Percent live tissue coverage was estimated to the nearest 5%.

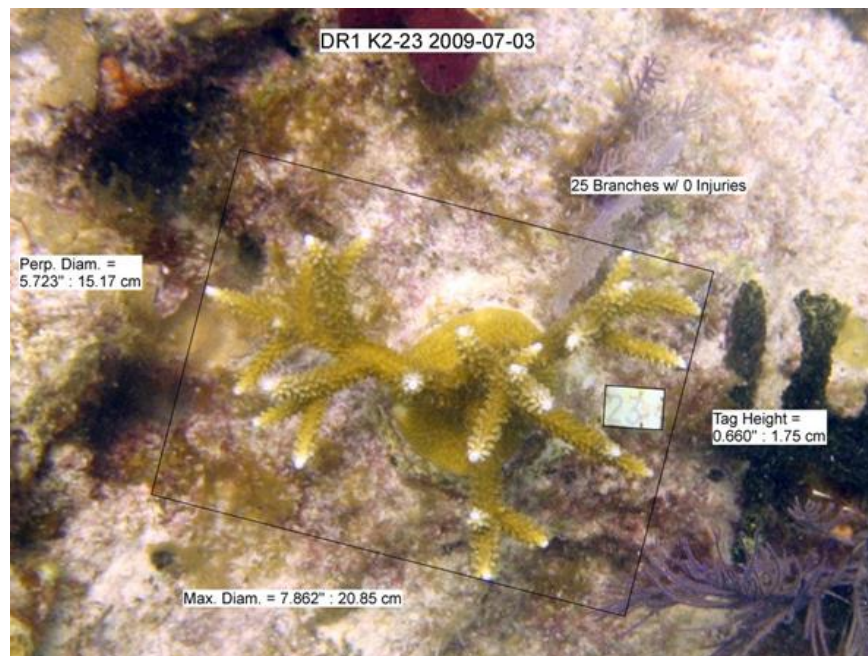


Figure 4. Example of a photograph used with CANVAS software. The image was taken on July 3, 2009 of a colony from the first project at Dry Rocks Ball #1 originally transplanted on November 12, 2008. The green rubber tag (23) corresponds to a K2 genotype and is 1.75 cm wide. The maximum and perpendicular skeletal diameters are extrapolated from the genotype tag size reference. Percent live tissue coverage was estimated to the nearest 5%.

Table 1. Composition of photographic record utilized in analyses obtained from the Coral Restoration Foundation organized by date of initial transplanting.

| Reef Name | Reef Type | Project ID & Initial Transplant Date | Genotypes Utilized | # Individual Colonies | # of Photos | Length of Record (yrs) |
|------------|---------------|--------------------------------------|---|-----------------------|-------------|------------------------|
| Molasses | Spur & Groove | ML12, 2007-07-26 | K1, K2, K3 | 18 | 298 | 6.98 |
| Pickles | Spur & Groove | PK1, 2008-07-20 | K1, K2, K3 | 18 | 297 | 6.28 |
| White Bank | Patch Reef | WB1, 2008-08-27 | K1, K2, K3 | 18 | 254 | 1.57 |
| Molasses | Hard-bottom | MD32, 2008-10-11 | K1, K2, K3 | 18 | 244 | 5.76 |
| Dry Rocks | Spur & Groove | DR1, 2008-11-12 | K1, K2, K3 | 18 | 184 | 5.77 |
| Molasses | Patch Reef | MT1, 2009-01-22 | K1, K2, K3 | 18 | 98 | 1.07 |
| French | Spur & Groove | FR1, 2009-04-02 | K1, K2, K3 | 18 | 96 | 5.42 |
| Dry Rocks | Spur & Groove | DR2, 2009-07-13 | K1, K2, K3 | 24 | 169 | 5.22 |
| Conch | Hard-bottom | CS1, 2009-08-04 | K1, K2, K3 | 24 | 181 | 1.52 |
| Conch | Hard-bottom | CS2, 2009-10-16 | K1, K2, K3 | 24 | 166 | 1.32 |
| Pickles | Hard-bottom | PK2, 2009-10-29 | K1, K2, K3 | 24 | 74 | 5.00 |
| French | Spur & Groove | FR2, 2010-05-27 | K1, K2, K3 | 24 | 71 | 4.75 |
| Molasses | Patch Reef | MT2, 2010-07-30 | K1, K2, K3 | 24 | 42 | 0.95 |
| Pickles | Hard-bottom | PKARRA, 2012-04-24 | U1, U3, U17, U44, U51, U53, U54, U55, U56, U59, U61 | 400 | 153 | 2.56 |
| Conch | Hard-bottom | CNARRA, 2012-05-12 | U1, U3, U17, U44, U51, U53, U54, U55, U57, U59, U61, U62, U63 | 400 | 150 | 2.67 |
| Molasses | Spur & Groove | MLARRA, 2012-05-22 | U1, U3, U17, U44, U51, U53, U54, U55, U59, U61 | 400 | 116 | 2.63 |

| | | | | | | |
|-----------|---------------|--------------------|---|-----|-----|------|
| Dry Rocks | Spur & Groove | DRARRA, 2012-06-05 | U1, U3, U17, U44, U51, U53, U54, U55, U56, U59, U61 | 400 | 129 | 2.63 |
| Pickles | Spur & Groove | P2013, 2013-08-16 | U7, U12, U15, U20, U21, U22, U24, U25, U28, U32, U37, U42, U59, U68, U69, U70 | 296 | 97 | 1.32 |
| French | Spur & Groove | FR2013, 2013-08-06 | U1, U4, U7, U12, U52, U53, U65, U69, U70, U79 | 161 | 35 | 1.07 |
| Conch | Hard-bottom | CNC, 2013-12-17 | U51, U52, U53, U54, U55, U56, U58, U59, U63, U64 | 92 | 20 | 1.10 |

In situ resampling for colony sizes and condition in 2014 using SCUBA was conducted to record the current status of the 17 projects used in the photographic analyses and to establish new baselines for these projects for future monitoring efforts.

Hypotheses for Objective #1 include:

H₀: No difference in growth, survivorship, or condition of nursery-raised *Acropora cervicornis* transplants based on location, genotype, or habitat.

H₁: Differences exist in growth, survivorship, or condition of nursery-raised *Acropora cervicornis* transplants, based on location, genotype, or habitat.

LITTLE CONCH REEF EXPERIMENT

In October and November, 2013, 1,288 *Acropora cervicornis* colonies were transplanted from the Coral Restoration Foundation Tavernier nursery to Little Conch Reef using CRF staff and CRF-trained volunteers. A transplant experiment was designed to evaluate the effects of transplant methodology and genotype on colony growth and survivorship at replicated natural densities. The colonies were placed at two depths (5m

and 12m), in either a cluster or a thicket configuration (Figures 5 and 6). Clusters are defined as a group of approximately 14 colonies arranged in a solid circular structure 1m in diameter. Thickets contain approximately 140 colonies in a 3.5m by 1.5m rectangular configuration. Six of the clusters at each depth contained one individual from each of 14 different genotypes (multigenetic clusters) while the remaining ten clusters were composed of a single genotype (monogenetic clusters). Each thicket contained all 14 genotypes, with each genotype comprising a subunit within the larger configuration (Figures 7 and 8). As in the photographic analysis, CRF records provided genotype information on the colonies.

The shallow (5m) site was located at 24°56.797' N, 80°28.157' W. The deep (12m) site was located approximately 350m to the northeast at 24°56.855' N, 80°27.968' W. Both sites are described as hard-bottom pavement sites (Florida Fish and Wildlife Conservation Commission 2014). The deep site had a ledge and sand channel to the east while the shallow site has a ledge and sand channel to the north. A natural *A. cervicornis* thicket is located approximately 30m to the southeast of the shallow site. No natural colonies were found at or close to the deep site.



Figures 5 and 6. Examples of a cluster (left) and thicket (right) utilized in the Little Conch experiment. Both images are from the shallow (5m) site (Cluster #11 and Thicket #1) taken on November 22, 2013.

On June 6, 2014, 963 colonies were added to the shallow site by stacking them on top of existing structures. The additional colonies were secured to the original colonies with plastic zip-ties, resulting in structures approximately 0.5m tall. Three multigenetic clusters, four monogenetic clusters, and one thicket were used for stacking. The original experimental design called for all of the clusters and thickets to receive additional corals for stacking. However, there were an insufficient number of colonies available from the nursery to achieve this result. In total, 2,251 colonies were transplanted for this study. The rationale for stacking was to simulate densities and topography typically found in natural staghorn thickets. This would evaluate if larger structures would survive better than small, individual colonies. Only the shallow site received additional colonies due to a significant disease-related mortality event that occurred almost immediately after the corals were transplanted at the deep site. The shallow site was not impacted by the disease event. Structures which received additional corals are termed “stacked” while those that did not are termed “non-stacked”. It was hoped that stacked colonies secured together by plastic zip-ties would fuse to create a solid framework.

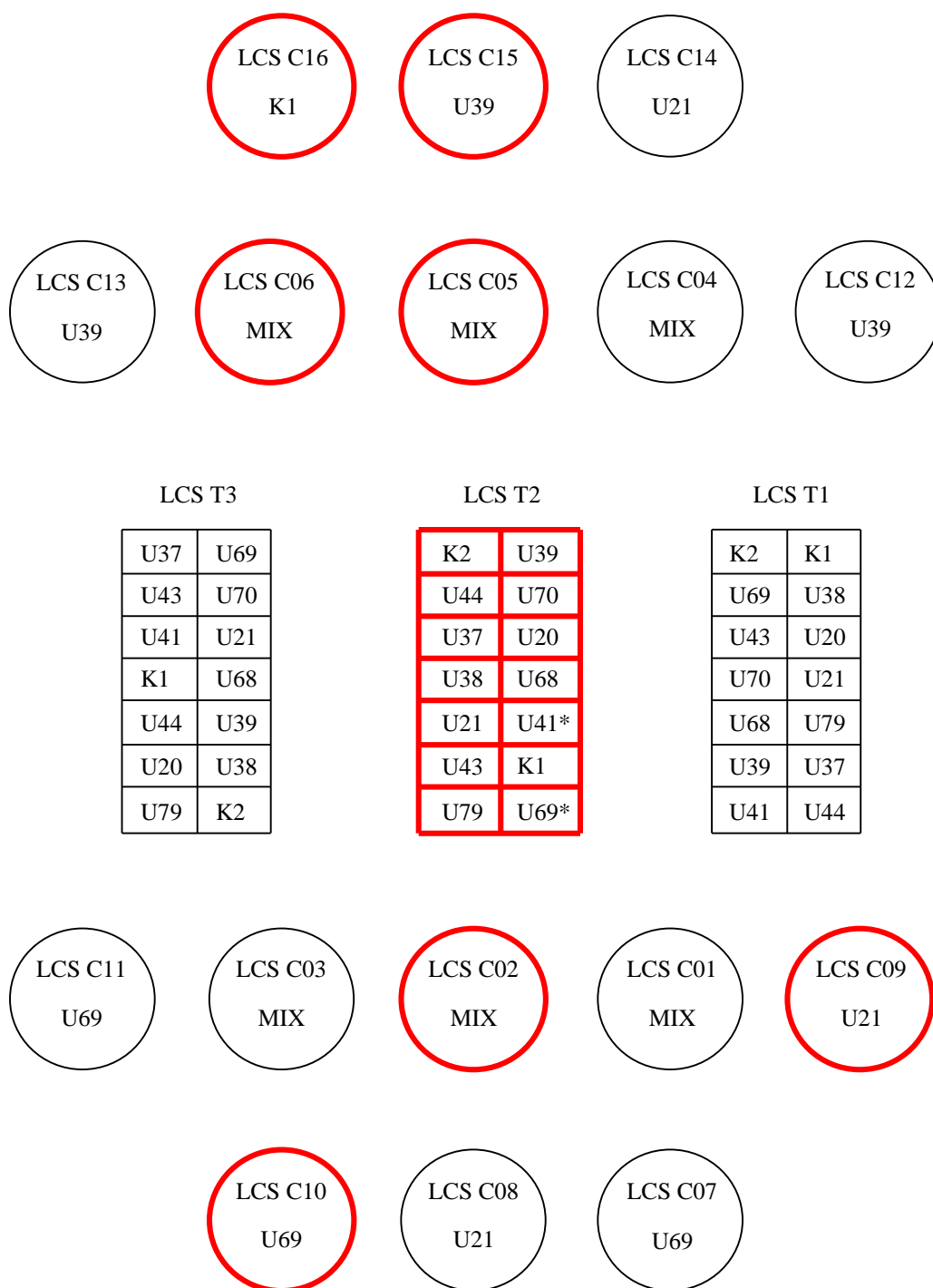


Figure 7. Map of the shallow (5m) Little Conch site. Circles represent clusters while rectangles represent thickets. Text indicates structural identifier and genetic composition. “MIX” indicates a multigenetic composition. Initially, 644 colonies were deployed between October 10 and November 2, 2013. 963 colonies were added on June 6, 2014. Structures which received additional corals are shown in red. * U41 and U69 in LCS T2 did not receive additional colonies due to limited supply from the nursery.

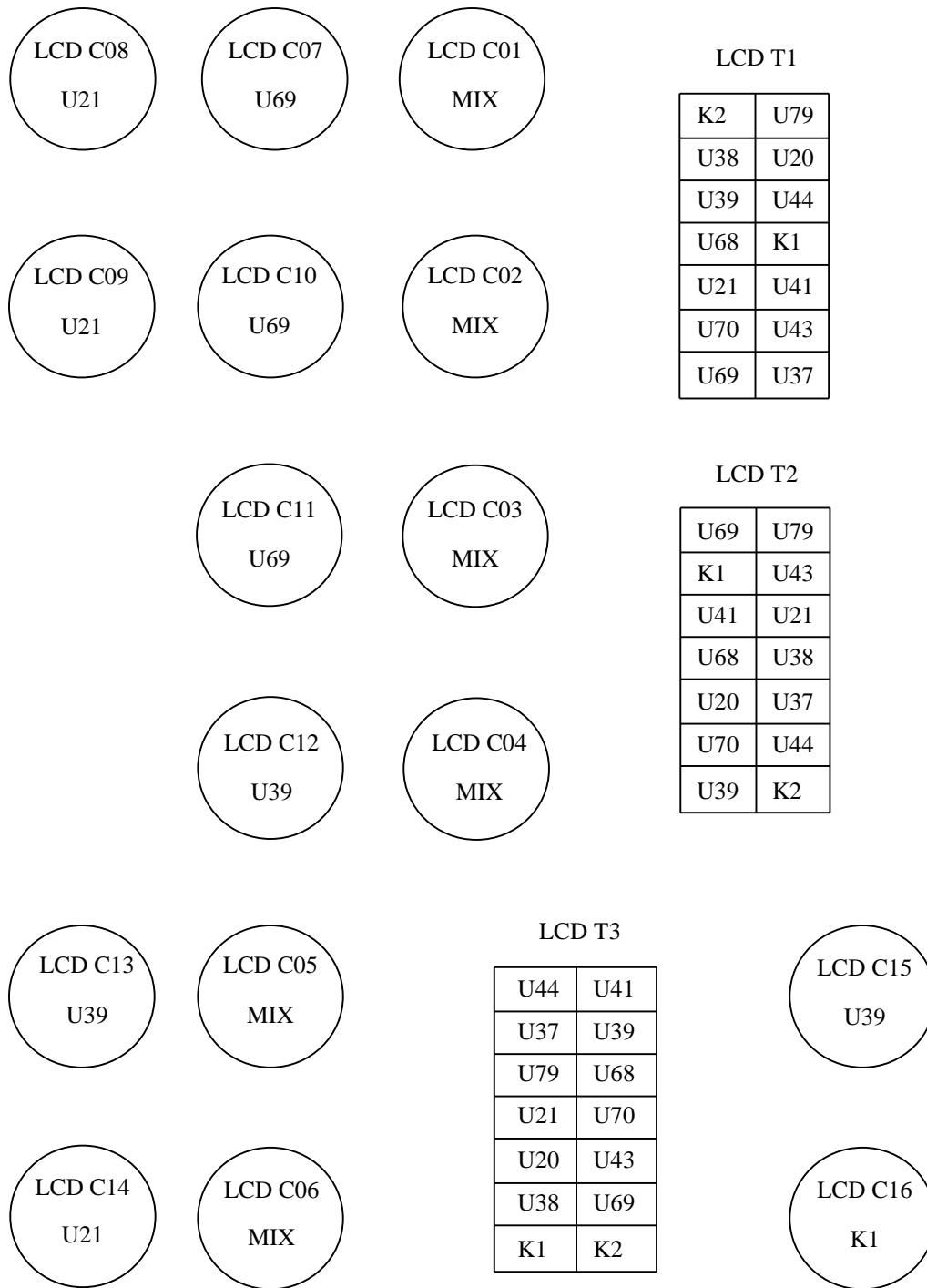


Figure 8. Map of the deep (12m) Little Conch site. Circles represent clusters while rectangles represent thickets. Text indicates structural identifier and genetic composition. “MIX” indicates a multigenetic composition. A total of 644 colonies were deployed between October 10 and November 2, 2013. This site did not receive additional corals in 2014 due to high disease mortality observed early in the project.

The transplant experiment was monitored *in situ* using SCUBA approximately every three months through January 2015. Colony maximum skeletal diameter, perpendicular diameter, height, percent live tissue, and condition were measured at each sampling interval. For the stacked structures, identifying individual colonies was impossible, so the dimensions of the entire structure were recorded, as well as an approximation of percent live tissue. The monitoring period included a significant disease event at the start of the experiment at the deep site, and a Keys-wide bleaching episode. Photo-mosaics of the sites were conducted by researchers from the University of Miami Rosenstiel School of Marine and Atmospheric Science at approximately the same times as the *in situ* sampling.

Hypotheses for Objective #2 are:

H₀: No difference in growth, survivorship, or condition of nursery-raised *Acropora cervicornis* transplants at Little Conch Reef, based on depth, transplant structure, or genetic composition.

H₁: Differences exist in growth, survivorship, or condition of nursery-raised *Acropora cervicornis* transplants at Little Conch Reef, based on depth, transplant structure, or genetic composition.

DATA ANALYSES

Maximum skeletal diameter, survivorship, and percent live tissue coverage from both projects were collated in Microsoft Excel. Data exploration revealed that the data was not normally distributed. Non-parametric Wilcoxon and Kruskal-Wallis tests were used to compare means and were executed using R (R Core Team 2014). Survival was treated as a categorical response so colonies were assigned a 1 if they contained any amount of living tissue and a 0 if no living tissue could be identified. For the photographic analyses, sample sizes from individual projects were often too small for meaningful analysis so projects were grouped together by the year in which they were initiated. Mean maximum skeletal diameter, survivorship, and condition were compared across reefs, genotype, and habitat type. For the Little Conch experiment, mean maximum skeletal diameter, survivorship, and percent live tissue coverage were

compared across depth, transplant structure, and genetic composition. Stacked structures were compared separately from the non-stacked structures.

RESULTS

HISTORICAL PHOTOGRAPHIC ANALYSES

The results of 17 past Coral Restoration Foundation projects are included below, presented by the year the project began. They cover a seven-year time span, six different reefs, and 2,428 nursery-raised *Acropora cervicornis* colonies.

2007 PROJECT

On July 26, 2007, Coral Restoration Foundation (CRF) deployed 18 *Acropora cervicornis* colonies under Molasses Reef Ball #12 at a depth of 7m (project ID “ML12”). The transplants included three genotypes (K1, K2, and K3), with six representatives for each. They were arranged using a triangular configuration with rubber genotype tags epoxied to the base of the colonies. CRF conducted eight visits to this site between 2007 and 2010. A final *in situ* measurement was taken on July 16, 2014. This represents the longest single record in this study.

The average (\pm SE) initial maximum skeletal diameter for the 18 colonies was 9.54 ± 1.02 cm. By the last CRF visit on August 8, 2010, the average maximum skeletal diameter had increased to 40.84 ± 2.12 cm (Figure 9). The average size recorded in 2014 was similar to 2010 at 40.75 ± 10.80 cm, but with much higher variance.

Maximum Skeletal Diameter vs. Time

Molasses Reef Ball #12 (2007)

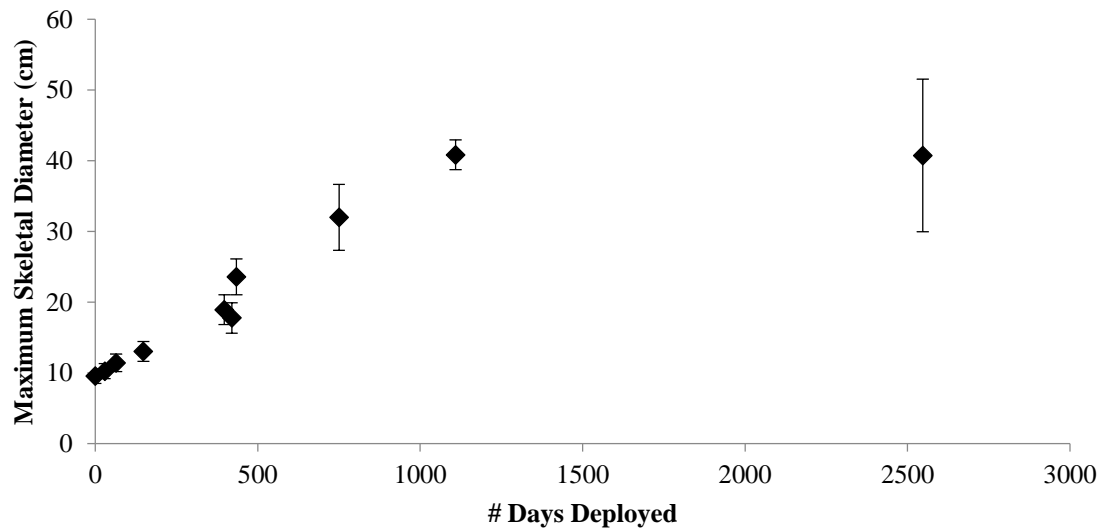


Figure 9. Maximum skeletal diameter vs. number of days deployed for Molasses Reef #12 (2007).

Totals = 2007-07: 18 colonies, 2007-08: 17, 2007-09: 18, 2007-12: 18, 2008-08: 18, 2008-09: 16, 2008-10: 11, 2009: 11, 2010: 3, 2014: 4

Maximum Depth: 7m

Error bars indicate standard error.

Each genotype displayed similar maximum diameters until 2014. K1 and K3 both recorded smaller sizes than their 2010 measurements, whereas K2 continued to increase (Figure 10). In 2014, K2 measured 70.00 cm across (n=1), K3 was 36.00 cm (n=1), and K1 averaged 28.50 ± 10.50 cm (n=2).

Maximum Skeletal Diameter vs. Time By Genotype

Molasses Reef Ball #12 (2007)

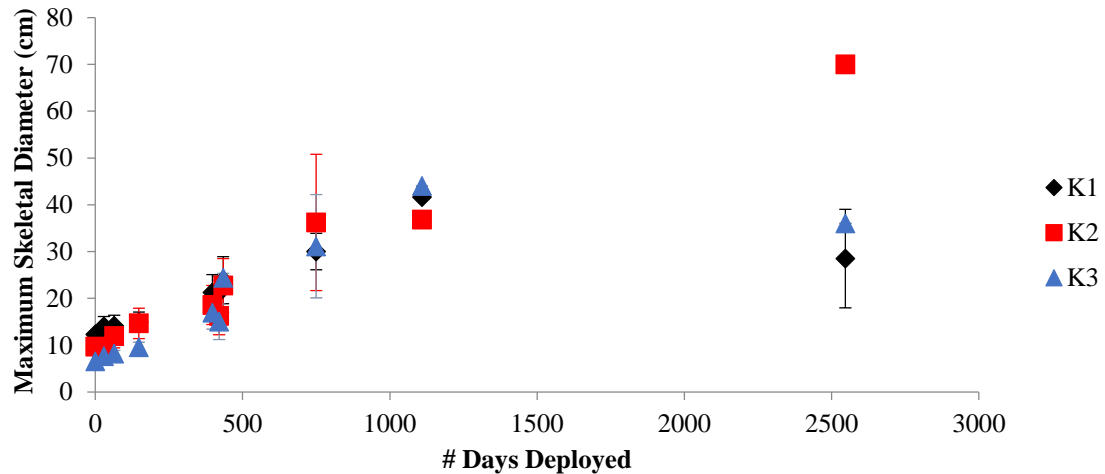


Figure 10. Maximum skeletal diameter vs. number of days deployed separated by genotype for Molasses Reef #12 (2007).

Totals =

K1 – 2007-07: 6 colonies, 2007-08: 6, 2007-09: 6, 2007-12: 6, 2008-08: 6, 2008-09: 6, 2008-10: 4, 2009: 5, 2010: 1, 2014: 2

K2 – 2007-07: 6 colonies, 2007-08: 5, 2007-09: 6, 2007-12: 6, 2008-08: 6, 2008-09: 5, 2008-10: 4, 2009: 3, 2010: 1, 2014: 1

K3 – 2007-07: 6 colonies, 2007-08: 6, 2007-09: 6, 2007-12: 6, 2008-08: 6, 2008-09: 5, 2008-10: 3, 2009: 3, 2010: 1, 2014: 1

Maximum Depth: 7m

Error bars indicate standard error.

Mortality did not occur among any of the genotypes until the second year of the study. Between August 2008 and August 2009, six of the original 18 colonies died (Figure 11). During the cold water event in early 2010 (Lirman et al. 2011), the number of surviving colonies fell to just three. In both 2009 and 2010, a dichotomous distribution developed where colonies appeared either healthy or completely dead (Figure 12). By 2014, only two of the original colonies had any remaining live tissue. Small sample sizes prevented any statistical comparisons.

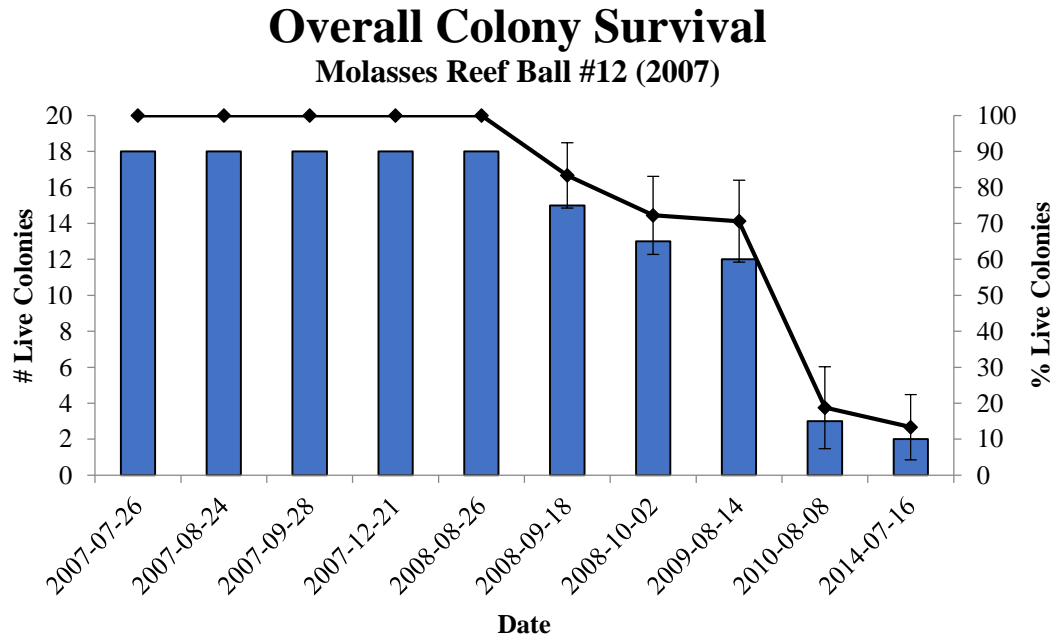


Figure 11. Colony survivorship by date for Molasses Reef #12 (2007).
 Bars indicate number of surviving colonies, regardless of percent live tissue.
 Line indicates percent survivorship ($(\# \text{ alive}/\text{total}) \times 100$)
 Totals = 2007-07: 18 colonies, 2007-08: 18, 2007-09: 18, 2007-12: 18, 2008-08: 18,
 2008-09: 18, 2008-10: 18, 2009: 17, 2010: 16, 2014: 15
 Maximum Depth: 7m
 Error bars indicate standard error.

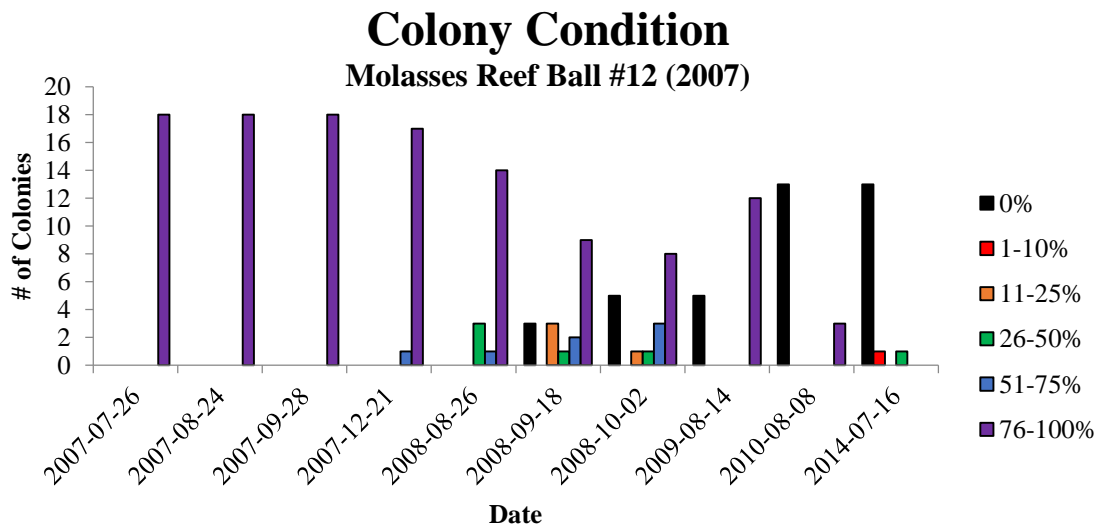


Figure 12. Histogram of colony condition by date for Molasses Reef #12 (2007).
 Totals = 2007-07: 18 colonies, 2007-08: 18, 2007-09: 18, 2007-12: 18, 2008-08: 18,
 2008-09: 18, 2008-10: 18, 2009: 17, 2010: 16, 2014: 15
 Maximum Depth: 7m

2008 PROJECTS

In 2008, four projects were followed – one each at Pickles Reef, White Bank, Molasses Reef, and Dry Rocks. For each project, 18 colonies were deployed in the triangular configuration using six representatives each from the K1, K2, and K3 genotypes. Rubber genotype tags were epoxied to the base of the colonies. Colonies at Pickles Reef (project ID “PK1”) were deployed on July 20, 2008 at a depth of 5m on a spur under Pickles Ball #1. The White Bank colonies (project ID “WB1”) were attached to a patch reef at 10m depth on August 27, 2008 near White Bank Ball #1. The Molasses Reef individuals (project ID “MD32”) were attached to a hard-bottom pavement habitat at a depth of 9m on October 11, 2008 under Molasses Ball #32. Lastly, the Dry Rocks project (project ID “DR1”) was transplanted on November 12, 2008 on a spur-and-groove structure at a depth of 9m under Dry Rocks Ball #1.

CRF conducted eight monitoring visits to Pickles Reef, six visits to White Bank, five to Molasses Reef, and four to Dry Rocks between 2008 and 2011. 2010 was the last year that all projects had an overlapping photographic record. *In situ* measurements were taken of Molasses Reef on July 16, 2014, Dry Rocks on August 19, 2014, and Pickles Reef on October 29, 2014. No genotype tags could be identified in the 2014 visit to Dry Rocks. No surviving colonies at White Bank were observed in 2014.

The average (\pm SE) initial maximum skeletal diameter for the 71 colonies transplanted was 9.10 ± 0.28 cm. All four sites were combined to assess the overall growth and survivorship of the transplanted colonies. In total, 72 colonies were transplanted but a photograph of one of the White Bank colonies was missing, resulting in an initial sample size of 71 colonies. By 2010, the average maximum skeletal diameter had increased to 38.85 ± 1.17 cm (Figure 13). The 2014 *in situ* measurements were similar to those in 2010 with an average diameter of 40.46 ± 3.96 cm. Difference between average maximum skeletal diameter in 2008 and 2010 was statistically significant (Wilcoxon test $p < 0.0001$). Difference between 2010 and 2014 was not significant.

Maximum Skeletal Diameter vs. Time

2008 Projects

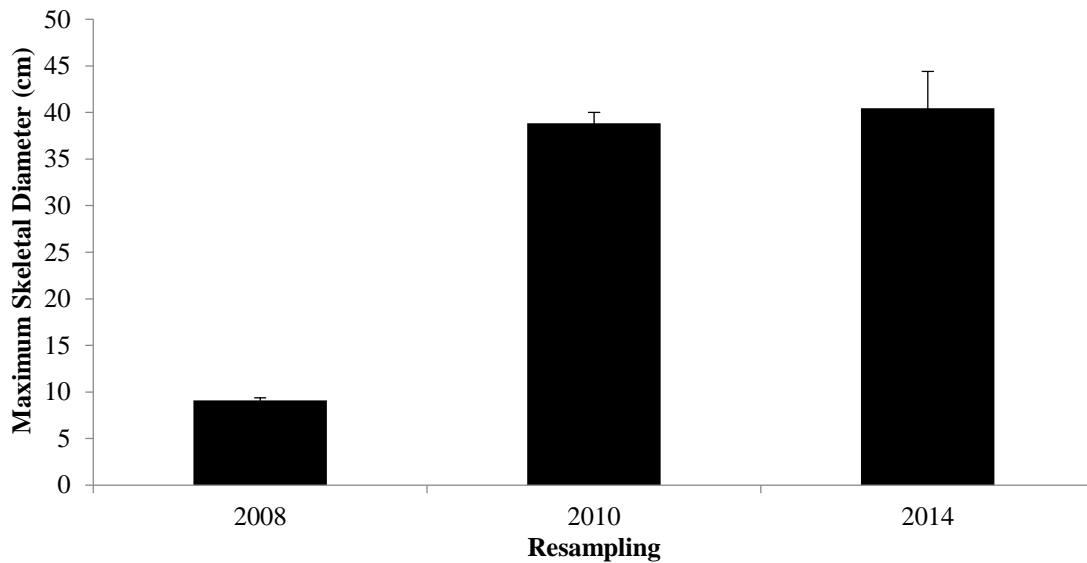


Figure 13. Maximum skeletal diameter by resampling for 2008 projects. Data averaged across all four reefs (Molasses Reef, Pickles Reef, White Banks, and Dry Rocks).
 Date Ranges = 2008: 7/20 - 11/12/2008, 2010: 3/18 - 8/6/2010, 2014: 7/16 - 10/29/2014
 Totals = 2008: 71 colonies, 2010: 51, 2014: 48
 Maximum Depth: 10m
 Error bars indicate standard error.

When analyzed by reef, no statistical difference was found in skeletal diameter within the sampling interval. In 2008, average maximum skeletal diameter was 10.26 ± 0.58 cm for Pickles Reef, 8.23 ± 0.42 cm for White Banks, 9.02 ± 0.44 cm for Molasses Reef, and 8.83 ± 0.69 cm for Dry Rocks. By 2014, average maximum skeletal diameter had increased to 35.67 ± 4.65 cm for Pickles Reef, 47.00 ± 6.12 cm for Molasses Reef, and 40.16 ± 5.14 cm for Dry Rocks (Figure 14). At its last recording in 2010, average skeletal diameter at White Bank was 30.73 ± 8.11 cm. Difference between average maximum skeletal diameter in 2008 and 2010 were statistically significant for all four reefs (Wilcoxon test $p \ll 0.0001$). Differences between 2010 and 2014 were not significant.

Maximum Skeletal Diameter vs. Time By Reef 2008 Projects

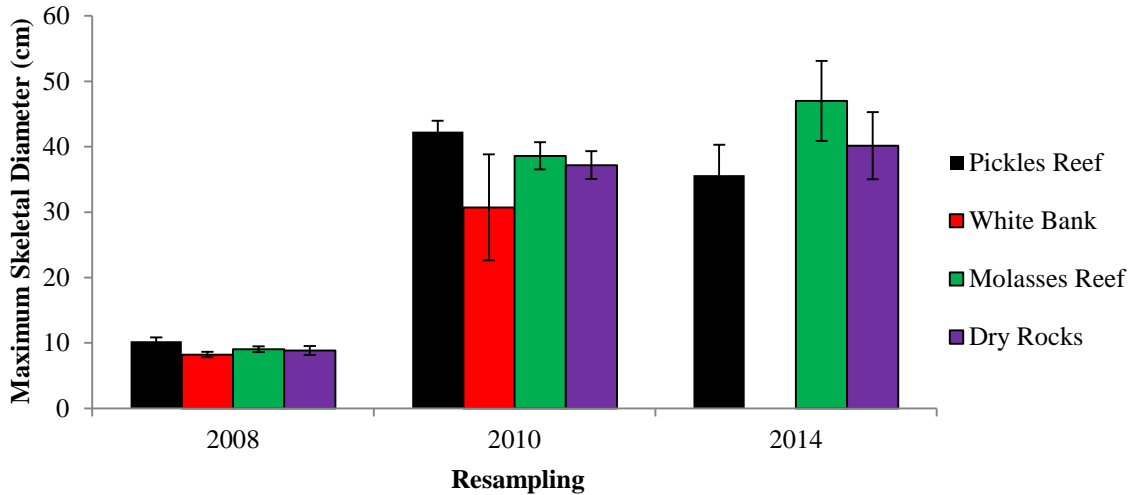


Figure 14. Maximum skeletal diameter by resampling separated by reef for 2008 projects. Date Ranges = 2008: 7/20 - 11/12/2008, 2010: 3/18 - 8/6/2010, 2014: 7/16 - 10/29/2014
Totals =
Pickles - 2008: 18 colonies, 2010: 14, 2014: 6, White Bank - 2008: 17, 2010: 2,
Molasses - 2008: 18, 2010: 18, 2014: 6, Dry Rocks - 2008: 18, 2010: 17, 2014: 36
Maximum Depth: 10m
Error bars indicate standard error.

When analyzed by genotype, K3 was initially smaller than K2 (8.18 ± 0.53 cm for K3, 9.43 ± 0.34 cm for K2, pairwise Wilcoxon test p-value 0.043 using Bonferroni adjustment). In 2008, K1 was the largest, but also had the highest standard error, preventing any statistical differentiation (9.64 ± 0.53 cm). No statistical difference existed in skeletal diameter among the three genotypes in 2010. By 2014, average maximum skeletal diameter had increased to 43.00 ± 5.90 cm for K1, 42.83 ± 7.23 cm for K2, and 33.50 ± 3.50 cm for K3 (Figure 15). Small sample sizes in 2014 prevented statistical comparisons for that year. Difference between average maximum skeletal diameter in 2008 and 2010 were statistically significant for all three genotypes (Wilcoxon test $p < 0.0001$). Differences between 2010 and 2014 were not significant.

Maximum Skeletal Diameter vs. Time By Genotype 2008 Projects

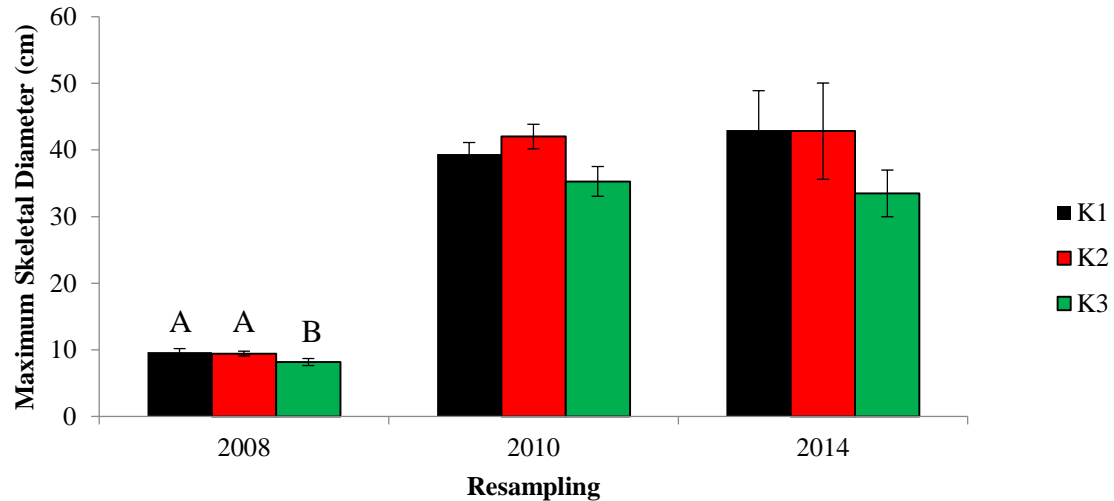


Figure 15. Maximum skeletal diameter by resampling separated by genotype for 2008 projects.

Date Ranges = 2008: 7/20 - 11/12/2008, 2010: 3/18 - 8/6/2010, 2014: 7/16 - 10/29/2014

Totals =

K1 - 2008: 24 colonies, 2010: 18, 2014: 4, K2 - 2008: 24, 2010: 16, 2014: 6,

K3 - 2008: 23, 2010: 17, 2014: 2

Maximum Depth: 10m

Error bars indicate standard error. Letters indicate statistical difference within sampling.

When analyzed by habitat type, no statistical difference in skeletal diameter was found (Figure 16). In 2008, average maximum skeletal diameter on the hard-bottom (Molasses Reef) site was 9.02 ± 0.44 cm, 8.23 ± 0.42 cm for the patch reef (White Bank), and 9.54 ± 0.46 cm for the spur-and-groove sites (Pickles Reef and Dry Rocks). By 2014, average maximum skeletal diameter had increased to 47.00 ± 6.12 cm for the hard-bottom and 39.52 ± 4.45 cm for the spur-and-groove. The patch reef habitat was excluded from analysis because of high mortality in 2010. Difference between average maximum skeletal diameter in 2008 and 2010 were statistically significant for both habitat types (Wilcoxon test $p < 0.0001$). Differences between 2010 and 2014 were not significant.

Maximum Skeletal Diameter vs. Time By Habitat Type 2008 Projects

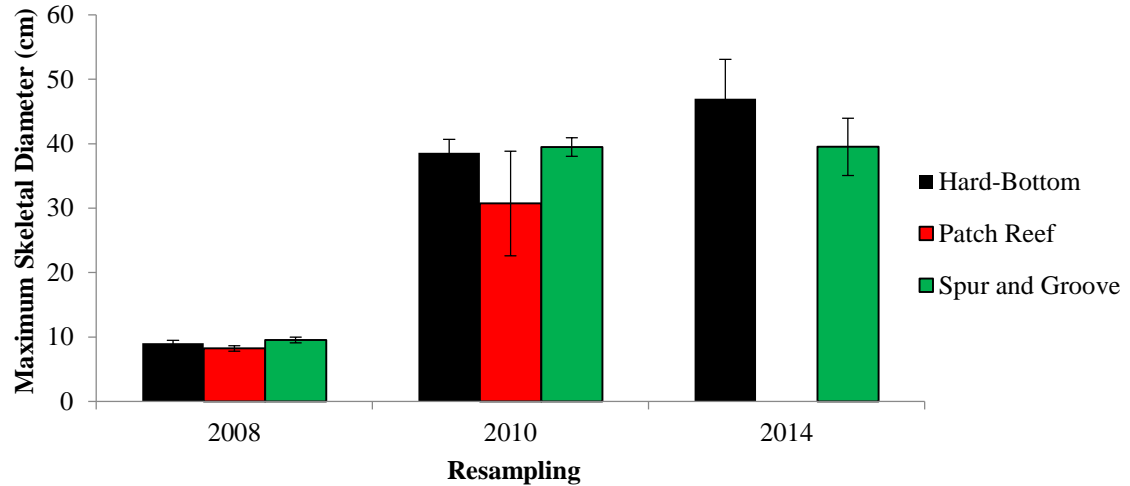


Figure 16. Maximum skeletal diameter by resampling separated by habitat type for 2008 projects.

Date Ranges = 2008: 7/20 - 11/12/2008, 2010: 3/18 - 8/6/2010, 2014: 7/16 - 10/29/2014

Totals =

Hard-Bottom - 2008: 18 colonies, 2010: 18, 2014: 6, Patch Reef - 2008: 17, 2010: 2,

Spur and Groove - 2008: 36, 2010: 31, 2014: 42

Maximum Depth: 10m

Error bars indicate standard error.

Survival and percent live tissue were variable across the four projects initiated in 2008. White Bank declined significantly following the 2010 cold water event ($11.11 \pm 7.62\%$ survival with an average live tissue coverage of $6.67 \pm 4.73\%$, Figure 17). Pickles Reef displayed less decline in 2010 ($83.33 \pm 9.04\%$ survival with $78.83 \pm 8.90\%$ live tissue, Figure 18). Molasses Reef and Dry Rocks were largely unaffected by the cold water event in 2010 (100% survival with $98.22 \pm 0.85\%$ live tissue, and 100% survival with $89.56 \pm 5.35\%$ live tissue, respectively). Differences in survival and percent live tissue at White Bank, relative to the other sites, were statistically significant (all pairwise Wilcoxon comparisons to White Bank $p \ll 0.001$ with Bonferroni adjustment). By 2014, colony survival at Molasses and Pickles Reefs were similar at $33.33 \pm 11.43\%$. Average percent live tissue at Molasses Reef was slightly higher than at Pickles Reef though the difference was not statistically significant ($13.89 \pm 6.49\%$ versus $11.06 \pm 6.55\%$).

Genotype tags at Dry Rocks could not be located during the 2014 *in situ* resampling, so

the results were excluded from the 2014 statistical comparison as the original colonies could not be separated from fragments.

Colonies at PK1 and MD32 recorded differences in percent survival between 2010 and 2014 (Wilcoxon $p < 0.01$), and between all three resamplings for percent live tissue (Wilcoxon $p < 0.05$). Percent survival and percent live tissue were both statistically different for WB1 between 2008 and 2010. Differences in percent survival between all three resamplings for DR1 were not statistically significant, but were significant for percent live tissue (Wilcoxon $p < 0.05$).

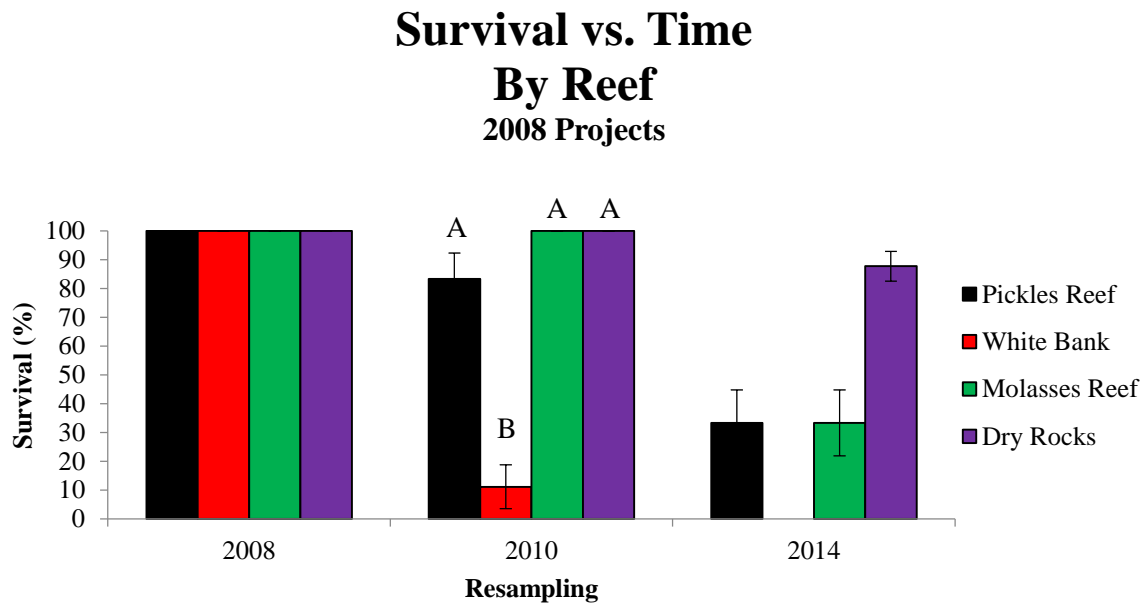


Figure 17. Colony survivorship by resampling separated by reef for 2008 projects.
 ((# alive/total)*100)
 Date Ranges = 2008: 7/20 - 11/12/2008, 2010: 3/18 - 8/6/2010, 2014: 7/16 - 10/29/2014
 Totals =
 Pickles - 2008: 18 colonies, 2010: 18, 2014: 18, White Bank - 2008: 17, 2010: 18,
 Molasses - 2008: 18, 2010: 18, 2014: 18, Dry Rocks - 2008: 18, 2010: 18, 2014: 41
 Maximum Depth: 10m
 Error bars indicate standard error. Letters indicate statistical significance within sampling.

Percent Live Tissue vs. Time By Reef 2008 Projects

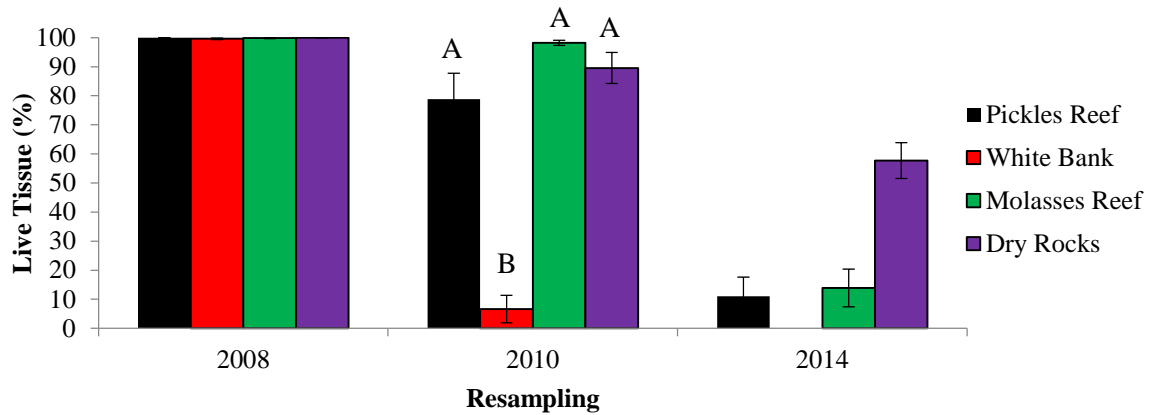


Figure 18. Percent live tissue by resampling separated by reef for 2008 projects.
 Date Ranges = 2008: 7/20 - 11/12/2008, 2010: 3/18 - 8/6/2010, 2014: 7/16 - 10/29/2014
 Totals =
 Pickles - 2008: 18 colonies, 2010: 18, 2014: 18, White Bank - 2008: 17, 2010: 18,
 Molasses - 2008: 18, 2010: 18, 2014: 18, Dry Rocks - 2008: 18, 2010: 18, 2014: 41
 Maximum Depth: 10m
 Error bars indicate standard error. Letters indicate statistical significance within sampling.

When analyzed by genotype, there were no statistical differences in survivorship or condition at any time interval (Figures 19 and 20). In 2010, percent survival had fallen to $79.17 \pm 8.47\%$ for K1, and $70.83 \pm 9.48\%$ for K2 and K3. Percent live tissue was reduced to $73.29 \pm 8.36\%$ for K1, $68.08 \pm 9.23\%$ for K2, and $63.58 \pm 9.32\%$ for K3. By 2014, percent survival had declined to $33.33 \pm 14.21\%$, $50.00 \pm 15.08\%$, and $16.67 \pm 11.24\%$ for K1, K2, and K3 respectively. Live tissue coverage showed a greater reduction than survival in 2014, as average live tissue fell to $11.42 \pm 6.9\%$ for K1, $10.17 \pm 5.78\%$ for K2, and $15.83 \pm 10.69\%$ for K3.

The difference in percent live tissue for all three genotypes was significantly different between all three resamplings (Wilcoxon $p < 0.01$). For K1, percent survival was significant between 2010 and 2014 (Wilcoxon $p < 0.05$). The difference in percent survival was statistically significant between 2008 and 2010 (Wilcoxon $p < 0.05$) but not between 2010 and 2014. All three resamplings were statistically different in percent survival for the K3 colonies (Wilcoxon $p < 0.05$).

Survival vs. Time By Genotype 2008 Projects

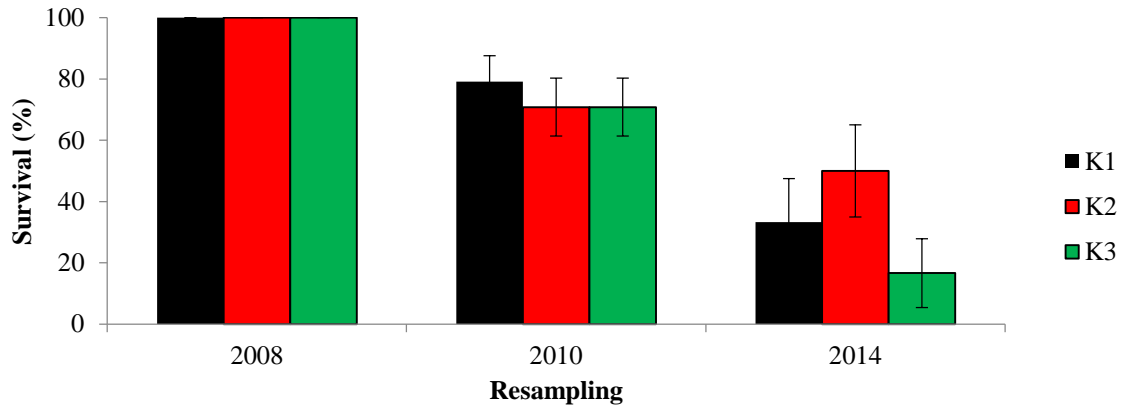


Figure 19. Colony survivorship by resampling separated by genotype for 2008 projects.
 ((# alive/total)*100)
 Date Ranges = 2008: 7/20 - 11/12/2008, 2010: 3/18 - 8/6/2010, 2014: 7/16 - 10/29/2014
 Totals = K1 - 24 colonies, 2010: 24, 2014: 12, K2- 2008: 24, 2010: 24, 2014: 12,
 K3 - 2008: 24, 2010: 24, 2014: 12
 Maximum Depth: 10m
 Error bars indicate standard error.

Live Tissue vs. Time By Genotype 2008 Projects

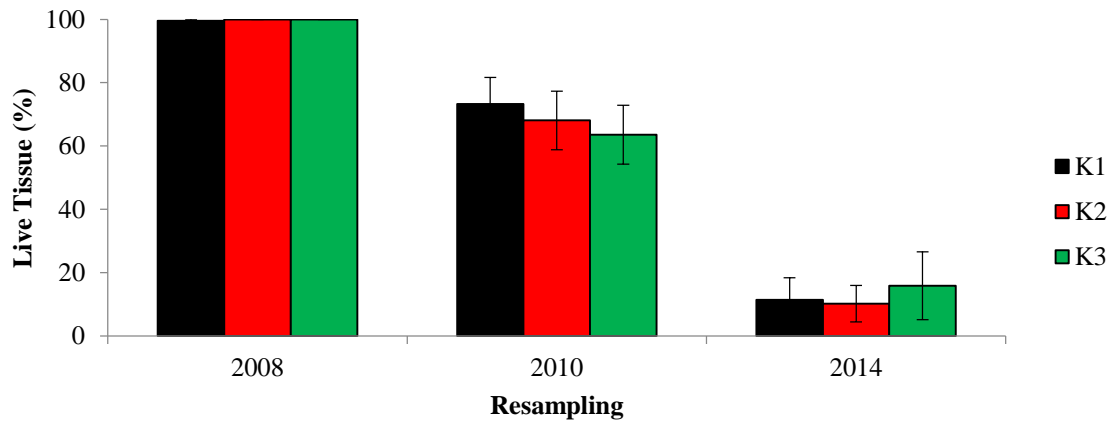


Figure 20. Percent live tissue by resampling separated by genotype for 2008 projects.
 Date Ranges = 2008: 7/20 - 11/12/2008, 2010: 3/18 - 8/6/2010, 2014: 7/16 - 10/29/2014
 Totals = K1 - 24 colonies, 2010: 24, 2014: 12, K2- 2008: 24, 2010: 24, 2014: 12,
 K3 - 2008: 24, 2010: 24, 2014: 12
 Maximum Depth: 10m
 Error bars indicate standard error.

When analyzed by habitat type, a statistically significant decline occurred in the patch reef habitat (White Bank) in 2010 relative to the hard-bottom (Molasses Reef, pairwise Wilcoxon $p \ll 0.0001$ with Bonferroni adjustment) and spur-and-groove habitats (Pickles Reef and Dry Rocks, pairwise Wilcoxon $p \ll 0.0001$ with Bonferroni adjustment). Following the 2010 cold water event, survivorship in the patch reef was $11.11 \pm 7.62\%$ and average percent live tissue was of $6.67 \pm 4.73\%$ (Figure 21). By comparison, the hard-bottom had 100% survival with $98.22 \pm 0.85\%$ live tissue, while the spur-and-groove sites had $91.67 \pm 4.67\%$ survival and $84.19 \pm 5.20\%$ live tissue (Figure 22). No difference was observed in 2014 between the hard-bottom and spur-and-groove habitats in either survival or percent live tissue. Measurements from the Dry Rocks spur-and-groove site were not included in the 2014 analysis due to lack of colony identification.

The differences in percent survival and percent live tissue for hard-bottom sites between 2008 and 2010 was not statistically significant, but was significant between 2010 and 2014 (Wilcoxon $p < 0.0001$). For spur-and-groove sites, percent survival was statistically different only between 2008 and 2014, while all of the time-steps were significantly different for percent live tissue (Wilcoxon $p < 0.0001$). The differences in percent survival and percent live tissue were statistically significant between 2008 and 2010 for colonies transplanted to the patch reef (Wilcoxon $p \ll 0.0001$).

Survival vs. Time By Habitat Type 2008 Projects

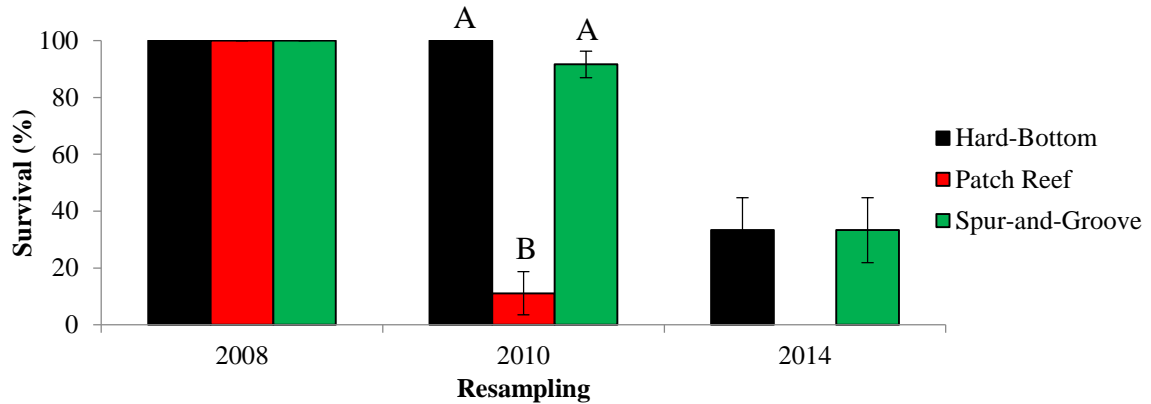


Figure 21. Colony survivorship by resampling separated by habitat type for 2008 projects.

((# alive/total)*100)

Date Ranges = 2008: 7/20 - 11/12/2008, 2010: 3/18 - 8/6/2010, 2014: 7/16 - 10/29/2014

Totals = Hard-Bottom - 2008: 18 colonies, 2010: 18, 2014: 18, Patch Reef - 2008: 18, 2010: 18, Spur-and-Groove - 2008: 18, 2010: 18, 2014: 18

Maximum Depth: 10m

Error bars indicate standard error. Letters indicate statistical difference within sampling.

Percent Live Tissue vs. Time By Habitat Type 2008 Projects

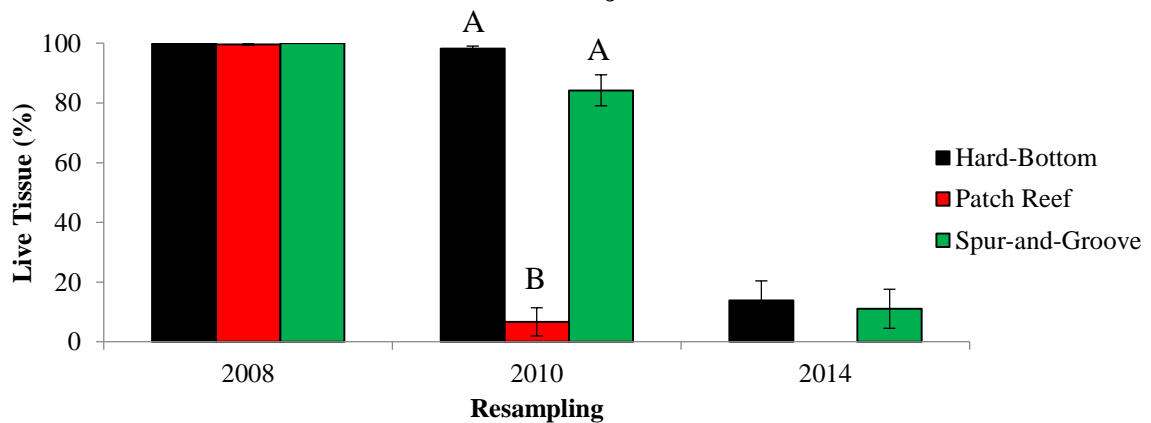


Figure 22. Percent live tissue by resampling separated by habitat type for 2008 projects.

Date Ranges = 2008: 7/20 - 11/12/2008, 2010: 3/18 - 8/6/2010, 2014: 7/16 - 10/29/2014

Totals = Hard-Bottom - 2008: 18 colonies, 2010: 18, 2014: 18, Patch Reef - 2008: 18, 2010: 18, Spur-and-Groove - 2008: 18, 2010: 18, 2014: 18

Maximum Depth: 10m

Error bars indicate standard error. Letters indicate statistical difference within sampling.

EARLY 2009 PROJECTS

To help clarify the analyses, projects were divided into “early 2009” (January through April) and “late 2009” (July through October) based on the original transplanting date. Two projects were followed during early 2009 – one at Molasses Reef and one at French Reef. For each project, 18 colonies were transplanted in the triangular configuration using six representatives each from the K1, K2, and K3 genotypes using rubber genotype tags epoxied to the base of the colonies. The Molasses Reef project (project ID “MT1”) began on January 22, 2009 at a depth of 8m on a patch reef within the Molasses Trench, 2.09km northwest from shallow spur-and-groove main reef. The French Reef colonies (project ID “FR1”) were transplanted on April 2, 2009 onto a spur-and-groove habitat at 9m depth under French Ball #6.

The Coral Restoration Foundation conducted three monitoring visits to MT1 and two to FR1 through 2010. An *in situ* visit to FR1 was conducted on November 2, 2014. No *in situ* visit to MT1 in 2014 was conducted due to high mortality recorded after the 2010 cold water event.

The average (\pm SE) initial maximum skeletal diameter for the 36 colonies deployed was 8.56 ± 0.33 cm. Both sites were combined to assess the overall growth and survivorship of the transplanted colonies. By 2010, the average maximum skeletal diameter had increased to 33.08 ± 1.77 cm (Figure 23). The 2014 *in situ* measurements from French Reef increased to an average diameter of 67.71 ± 6.04 cm. Differences in average maximum skeletal diameter between all three time-steps were statistically significant (Wilcoxon test $p \ll 0.0001$).

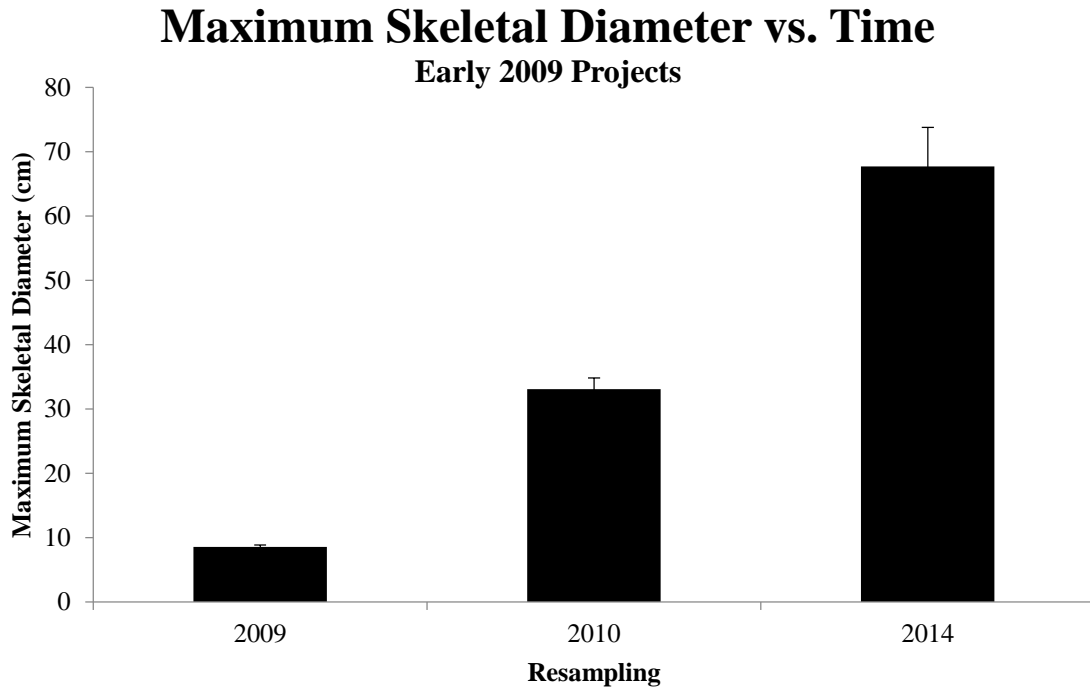


Figure 23. Maximum skeletal diameter by resampling for early 2009 projects. Data averaged across both reefs (Molasses Trench and French Reef). Date Ranges = 2009: 01/22 - 04/02/2009, 2010: 02/18 - 05/26/2010, 2014: 09/02/2014 Totals = 2009: 35 colonies, 2010: 16, 2014: 14 Maximum Depth: 9m Error bars indicate standard error.

When analyzed by reef, no statistical difference existed at the initial deployment in 2009. Low sample sizes at the Molasses site in 2010 prevented any further statistical comparison both within and between samplings. In 2009, average maximum skeletal diameter was 7.96 ± 0.46 cm for Molasses Reef, and 9.19 ± 0.43 cm for French Reef (Figure 24). In 2014, the average maximum skeletal diameter had increased to 67.71 ± 6.04 cm for French Reef. At its last recording in 2010, maximum skeletal diameter of the lone remaining colony at MT1 was 20.81 cm. Comparisons based on habitat type are identical to the comparison conducted by reef, where “MT1” can be replaced with “patch reef” and “FR1” with “spur-and-groove” (Figure 26). Differences in average maximum skeletal diameter between all three time-steps were statistically significant for French Reef (Wilcoxon test $p < 0.0001$).

Maximum Skeletal Diameter vs. Time By Reef

Early 2009 Projects

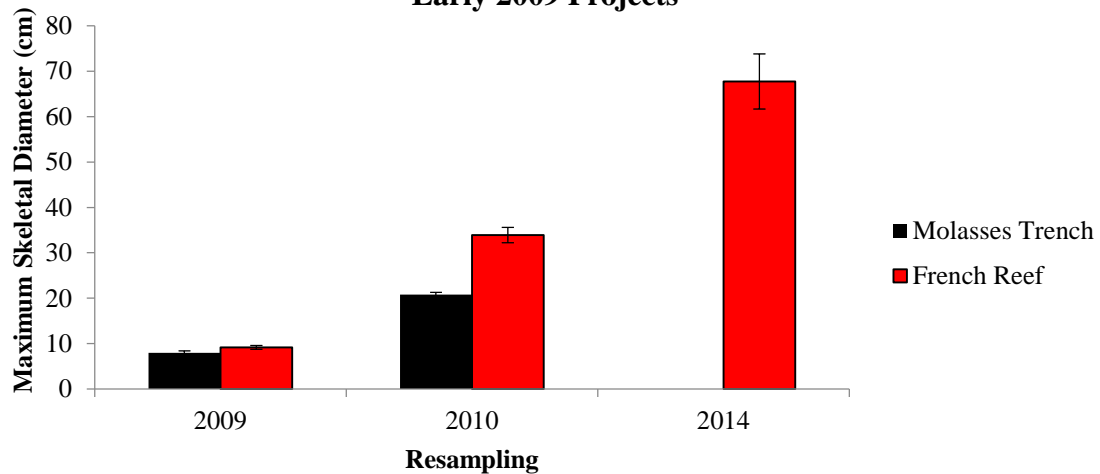


Figure 24. Maximum skeletal diameter by resampling separated by reef for early 2009 projects.

Date Ranges = 2009: 01/22 - 04/02/2009, 2010: 02/18 - 05/26/2010, 2014: 09/02/2014

Totals = Molasses Trench - 2009: 18 colonies, 2010: 1, 2014: 0,

French Reef - 2009: 17, 2010: 15, 2014: 14

Maximum Depth: 9m

Error bars indicate standard error.

When analyzed by genotype, K3 was initially smaller than K1 but no differences were identified in the 2010 or 2014 analyses (Kruskal-Wallis $p < 0.05$, pairwise Wilcoxon $p = 0.053$ with Bonferroni adjustment). In 2009, K1 colonies had an average maximum skeletal diameter of 9.35 ± 0.42 cm, while K2 and K3 recorded 8.68 ± 0.61 cm and 7.56 ± 0.57 cm, respectively (Figure 25). After just over one year on the reef, the average maximum skeletal diameter of the colonies had increased to 33.31 ± 2.38 cm for K1, 34.58 ± 2.84 cm for K2, and 29.09 ± 5.39 cm for K3. In 2014, the colonies had grown to 60.00 ± 6.56 cm for K1, 83.00 ± 16.02 cm for K2, and 63.20 ± 8.28 cm for K3.

Differences in average maximum skeletal diameter between 2009 and 2010 were statistically significant for K1 (Wilcoxon test $p < 0.001$), but not for 2010 and 2014 (Wilcoxon test $p = 0.052$). Differences in average maximum skeletal diameter for all three time-steps were significant for K2 (Wilcoxon test $p < 0.05$). Differences in average maximum skeletal diameter between 2009 and 2010 were statistically significant for K3 (Wilcoxon test $p < 0.05$), but not for 2010 and 2014 (Wilcoxon test $p = 0.11$).

Maximum Skeletal Diameter vs. Time By Genotype

Early 2009 Projects

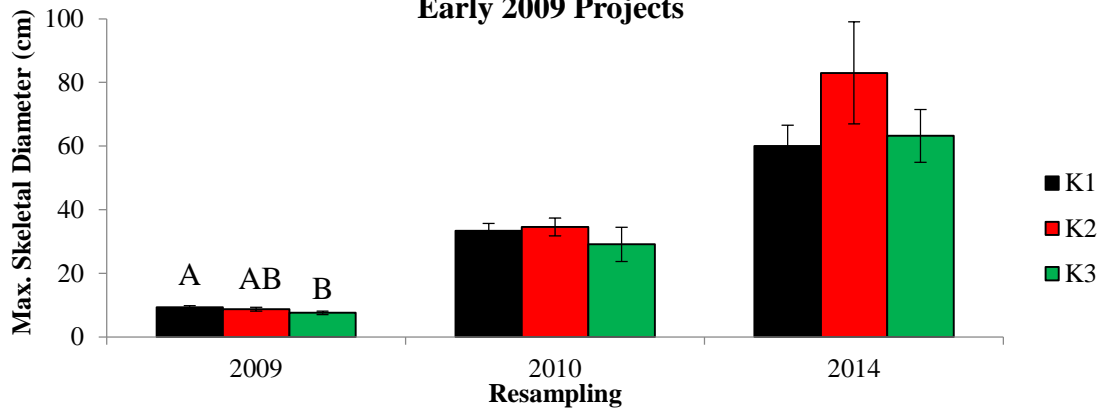


Figure 25. Maximum skeletal diameter by resampling separated by genotype for early 2009 projects.

Date Ranges = 2009: 01/22 - 04/02/2009, 2010: 02/18 - 05/26/2010, 2014: 09/02/2014

Totals = K1 - 2009: 12 colonies, 2010: 6, 2014: 5, K2 - 2009: 12, 2010: 7, 2014: 4, K3 - 2009: 12, 2010: 3, 2014: 5

Maximum Depth: 9m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

Maximum Skeletal Diameter vs. Time By Habitat Type

Early 2009 Projects

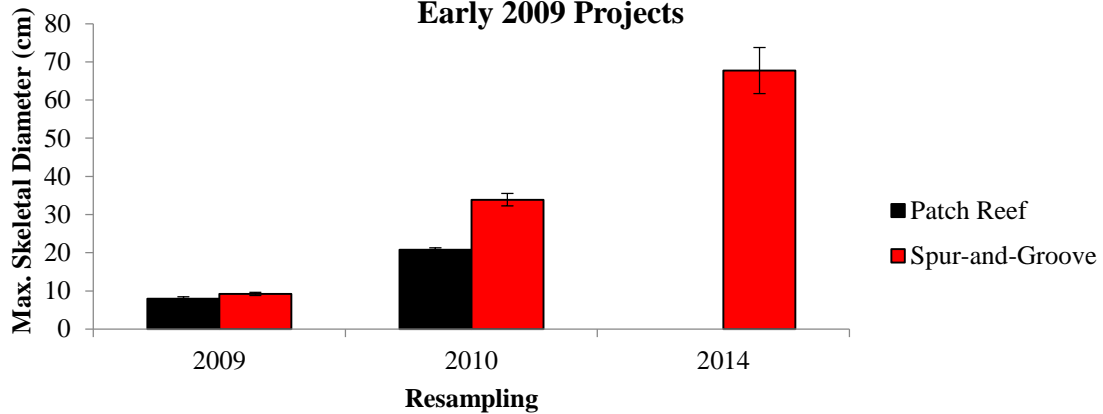


Figure 26. Maximum skeletal diameter by resampling separated by habitat type for early 2009 projects.

Date Ranges = 2009: 01/22 - 04/02/2009, 2010: 02/18 - 05/26/2010, 2014: 09/02/2014

Totals = Patch Reef - 2009: 18 colonies, 2010: 1, 2014: 0,

Spur-and-Groove - 2009: 17, 2010: 15, 2014: 14

Maximum Depth: 9m

Error bars indicate standard error

Percent survival and percent live tissue results were different between MT1 and FR1. Following the cold water event in early 2010, only a single colony out of the original 18 was observed alive at MT1 with 5% of its tissue remaining (Figures 26 and 27). At French Reef, 83.33 \pm 9.04% of the colonies survived with an average live tissue coverage of 83.33 \pm 9.04%. Differences in survival and condition were statistically significant (Wilcoxon $p < 0.0001$ for both). In 2014, FR1 still had 77.78 \pm 10.08% of its original colonies alive, with 22.83 \pm 5.20% live tissue. No follow-up visit was performed at MT1. The differences between 2009 and 2010 for percent survival and percent live tissue for Molasses Trench were statistically significant (Wilcoxon $p < 0.0001$). For French Reef, none of the time-steps were significantly different in percent survival. The difference between 2010 and 2014 were significantly different for percent live tissue (Wilcoxon $p < 0.001$) but between 2009 and 2010 was not. As in the skeletal diameter analysis, comparison based on habitat type for survival or condition is identical to the above comparison, where “MT1” and “FR1” with “patch reef” and “spur-and-groove”, respectively (Figures 31 and 32).

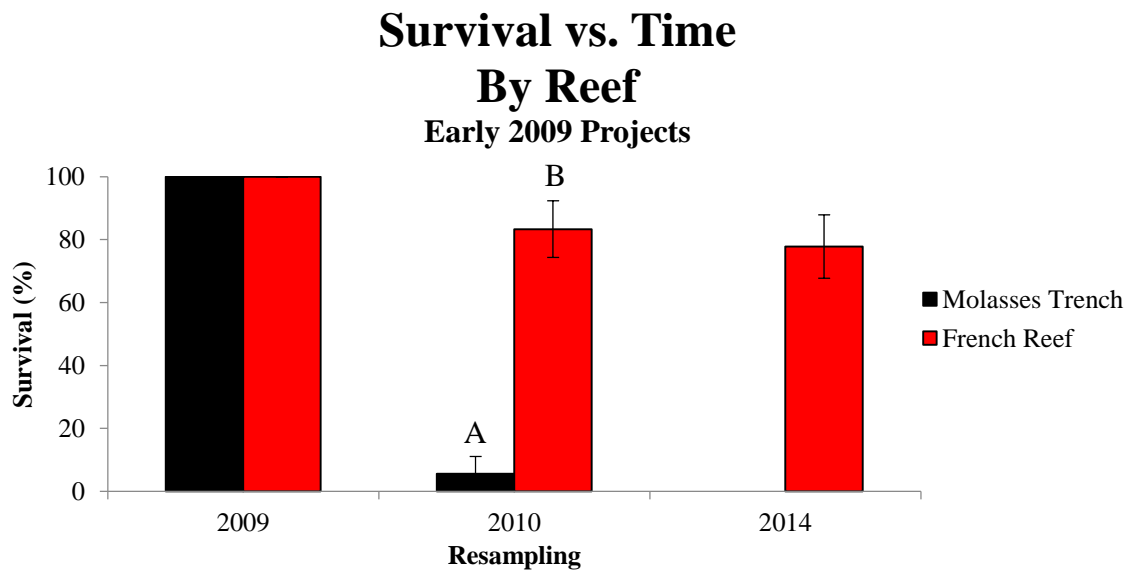


Figure 27. Colony survivorship by resampling separated by reef for early 2009 projects. ((# alive/total)*100)

Date Ranges = 2009: 01/22 - 04/02/2009, 2010: 02/18 - 05/26/2010, 2014: 09/02/2014

Totals = Molasses Trench - 2009: 18 colonies, 2010: 18, 2014: 0,

French Reef - 2009: 18, 2010: 18, 2014: 18

Maximum Depth: 9m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

Percent Live Tissue vs. Time By Reef Early 2009 Projects

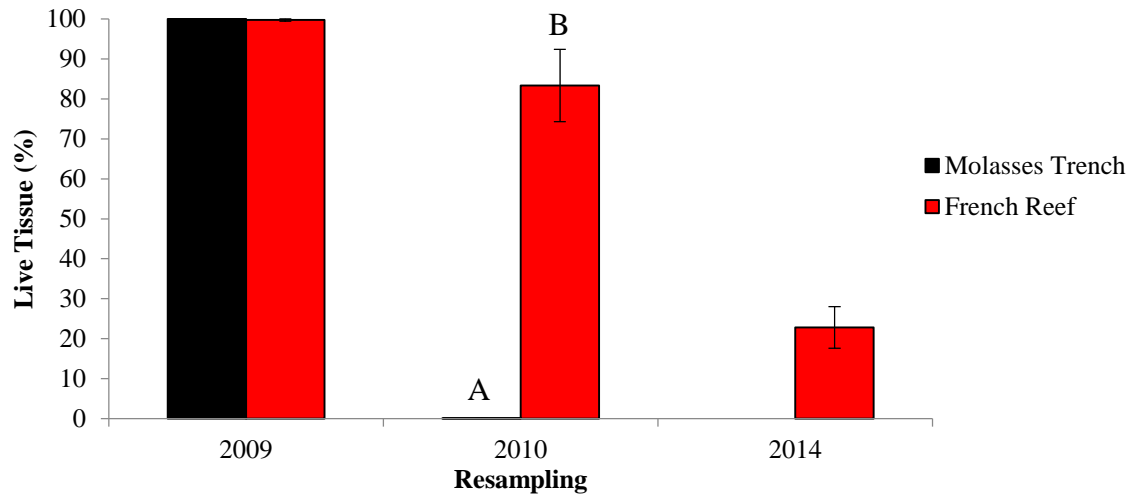


Figure 28. Percent live tissue by resampling separated by reef for early 2009 projects. Date Ranges = 2009: 01/22 - 04/02/2009, 2010: 02/18 - 05/26/2010, 2014: 09/02/2014
Totals = Molasses Trench - 2009: 18 colonies, 2010: 18, 2014: 0,
French Reef - 2009: 18, 2010: 18, 2014: 18
Maximum Depth: 9m
Error bars indicate standard error. Letters indicate statistical significance within sampling.

No statistical difference were seen among genotypes for either percent survival or percent live tissue. After the 2010 cold water event, $50.00 \pm 15.08\%$ of the K1 colonies, $58.33 \pm 14.86\%$ of the K2 colonies, and $25.00 \pm 13.06\%$ of the K3 colonies survived, with mortality highest at the Molasses site (Figure 28). Average percent live tissue coverage was $50.00 \pm 15.08\%$ for K1, $50.08 \pm 15.05\%$ for K2, and $25.00 \pm 13.06\%$ for K3 (Figure 29). By the 2014 *in situ* resampling at French Reef, $83.33 \pm 16.67\%$ of the K1 and K3 colonies were still alive, along with $66.67 \pm 21.08\%$ of the K2 colonies. Live tissue coverage had declined to $27.50 \pm 10.23\%$ for K1, $16.83 \pm 7.53\%$ for K2, and $24.17 \pm 10.12\%$ for K3 individuals. The Molasses site was excluded from the 2014 analysis due to nearly complete mortality in 2010. The differences in percent survival between 2009 and 2010 for all three genotypes was statistically significant (Wilcoxon $p < 0.05$). Percent survival for any genotype was not significant between 2010 and 2014 or 2009 and 2014.

The differences in percent live tissue between all three time-steps for all three genotypes were statistically significant (Wilcoxon $p < 0.05$).

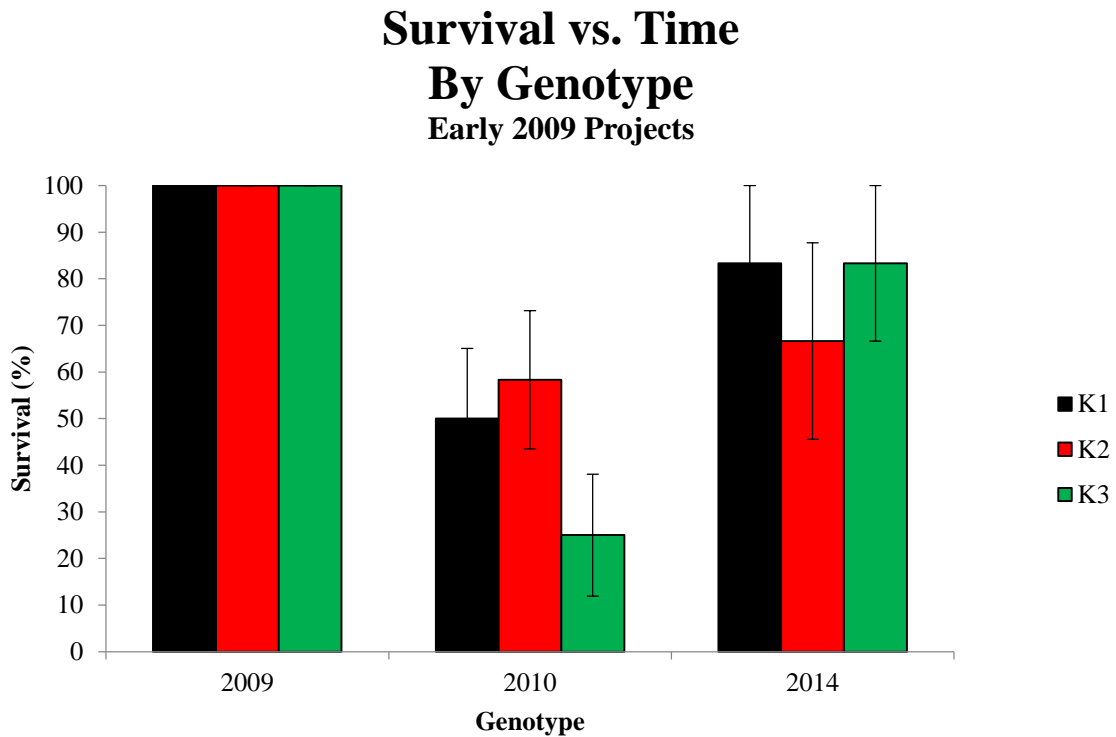


Figure 29. Colony survivorship by resampling separated by genotype for early 2009 projects.
Date Ranges = 2009: 01/22 – 04/02/2009, 2010: 02/18 - 05/26/2010, 2014: 09/02/2014
Totals = K1 – 2009: 12, 2010: 12, 2014: 6; K2 – 2009: 12, 2010: 12, 2014: 6;
K3 – 2009: 12, 2010: 12, 2014: 6
Maximum Depth: 9 m
Error bars indicate standard error.
2014 analysis is of FR1 only.

Percent Live Tissue vs. Time By Genotype

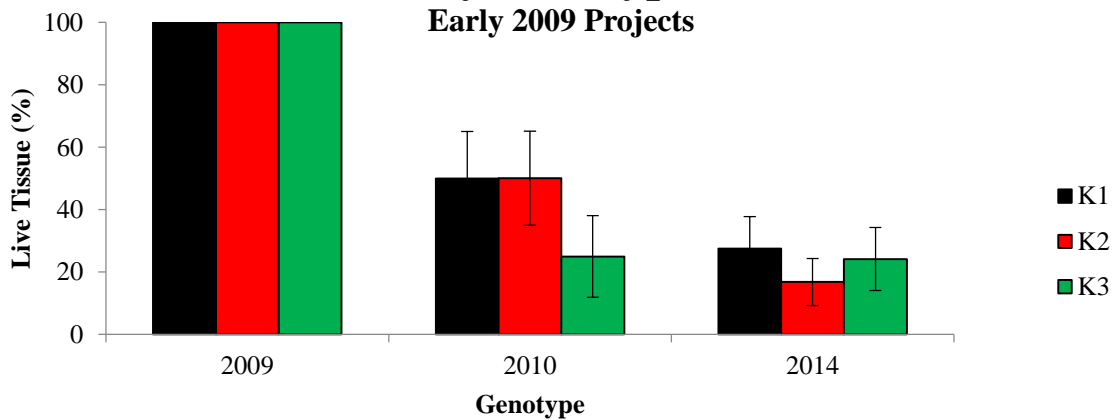


Figure 30. Percent live tissue by resampling separated by genotype for early 2009 projects.

Date Ranges = 2009: 01/22 – 04/02/2009, 2010: 02/18 - 05/26/2010, 2014: 09/02/2014

Totals = K1 – 2009: 12, 2010: 12, 2014: 6; K2 – 2009: 12, 2010: 12, 2014: 6;

K3 – 2009: 12, 2010: 12, 2014: 6

Maximum Depth: 9m

Error bars indicate standard error.

2014 analysis is of FR1 only.

Survival vs. Time By Habitat Type

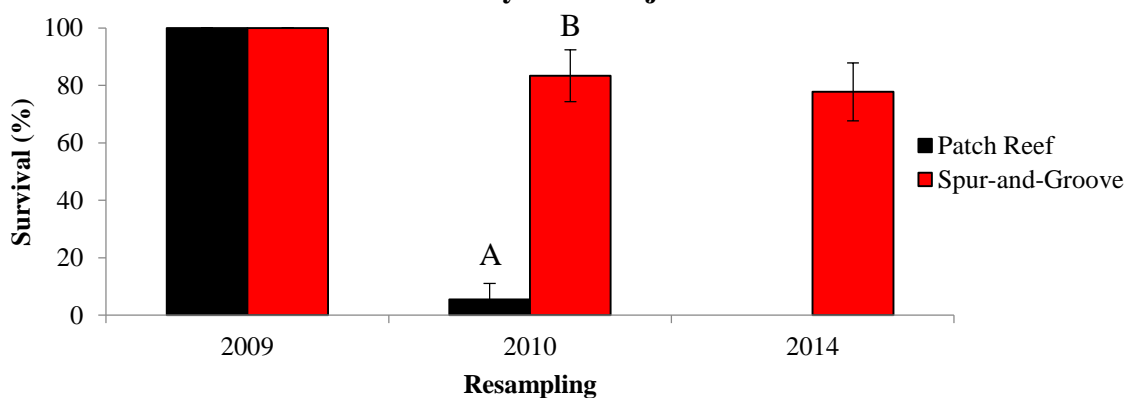


Figure 31. Colony survivorship by resampling separated by habitat type for early 2009 projects.

((# alive/total)*100)

Date Ranges = 2009: 01/22 - 04/02/2009, 2010: 02/18 - 05/26/2010, 2014: 09/02/2014

Totals = Patch Reef - 2009: 18 colonies, 2010: 18, 2014: 0,

Spur-and-Groove - 2009: 18, 2010: 18, 2014: 18

Maximum Depth: 9m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

Percent Live Tissue vs. Time By Habitat Type Early 2009 Projects

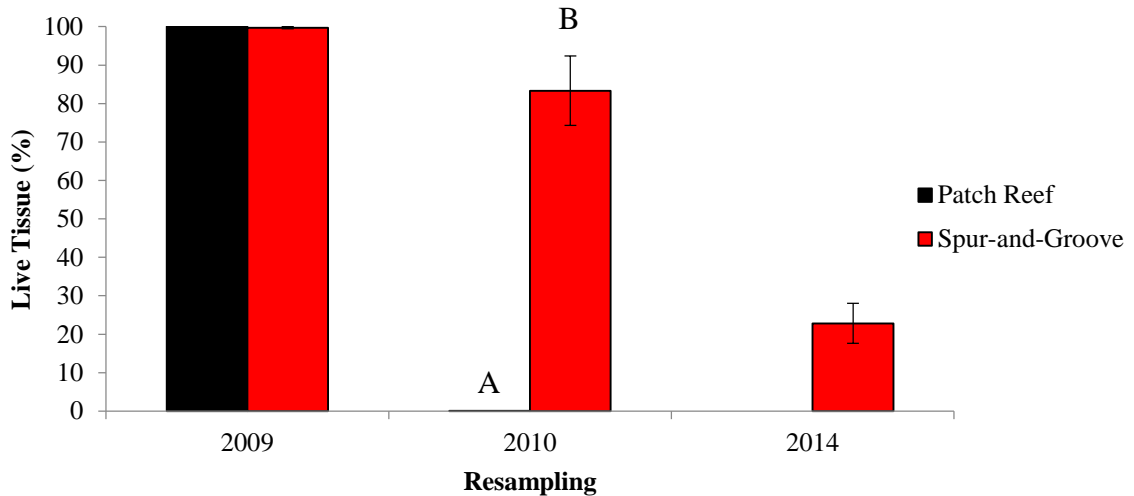


Figure 32. Percent live tissue by resampling separated by habitat type for early 2009 projects.

Date Ranges = 2009: 01/22 - 04/02/2009, 2010: 02/18 - 05/26/2010, 2014: 09/02/2014

Totals = Patch Reef - 2009: 18 colonies, 2010: 18, 2014: 0,

Spur-and-Groove - 2009: 18, 2010: 18, 2014: 18

Maximum Depth: 9m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

LATE 2009 PROJECTS

The “late 2009” projects were transplanted from July through October 2009. Four projects are in this category – one at Dry Rocks, two at Conch Reef, and one at Pickles Reef. The projects included 24 corals each transplanted in the triangular configuration using eight colonies each from the K1, K2, and K3 genotypes. Rubber genotype tags were epoxied to the base of the colonies. The Dry Rocks project (project ID “DR2”) began on July 13, 2009 on a spur-and-groove structure at a maximum depth of 5m under Dry Rocks Ball #8. The Conch Reef projects (project IDs “CS1” and “CS2”) were transplanted on August 4 and October 16, 2009, respectively. Both sites are on hard-bottom but at different depths – CS1 is at 8m while CS2 is at 5m. The Pickles Reef project (project ID “PK2”) began on October 29, 2009. It is a hard-bottom site at 9m depth under Pickles Ball #2.

CRF conducted three monitoring visits to DR2, CS1, and PK2 through 2011. Only two visits were conducted to CS2. *In situ* monitoring was conducted at DR2 on October 1, 2014 and at PK2 on October 29, 2014. No *in situ* visits were conducted at CS1 or CS2 in 2014 due to logistical challenges.

The average (\pm SE) initial maximum skeletal diameter for the 96 colonies transplanted was 9.23 ± 0.34 cm. By 2011, the average maximum skeletal diameter increased to 29.28 ± 1.17 cm (Figure 30). The 2014 *in situ* measurements increased to an average diameter of 54.59 ± 6.00 cm. Differences in average maximum skeletal diameter between all three time-steps were statistically significant (Wilcoxon test $p < 0.0001$).

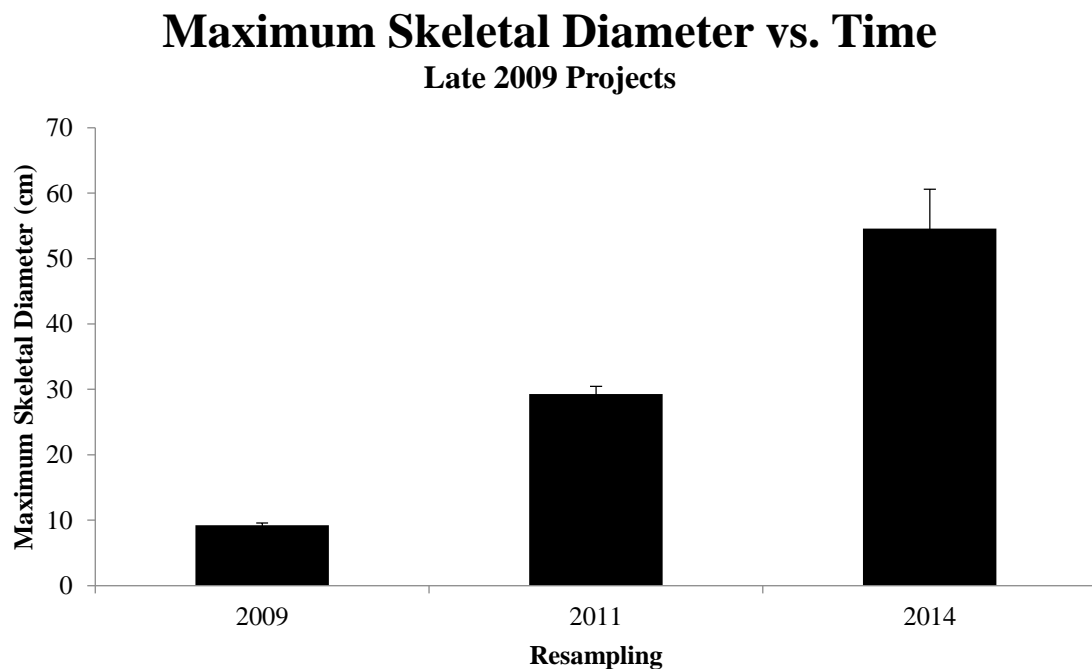


Figure 33. Maximum skeletal diameter by resampling for late 2009 projects. Data averaged across all three reefs (Conch Reef, Pickles Reef, and Dry Rocks). Date Ranges = 2009: 07/13 - 10/29/2009, 2011: 12/10/2010 - 6/15/2011, 2014: 10/01 - 10/29/2014
Totals = 2009: 96 colonies, 2011: 77, 2014: 17
Maximum Depth: 9m
Error bars indicate standard error.

When analyzed by reef, no statistical difference existed in maximum skeletal diameter at the initial deployment in 2009. In 2009, average maximum skeletal diameter was 9.70 ± 0.48 cm for Conch Reef, 8.63 ± 0.67 cm for Dry Rocks, and 8.89 ± 0.67 cm

for Pickles Reef (Figure 31). In 2011, the average maximum skeletal diameters increased to 25.16 ± 1.40 cm for Conch Reef, 38.64 ± 2.60 cm for Dry Rocks, and 26.72 ± 0.98 cm for Pickles Reef. The difference between Dry Rocks and the other two reefs was statistically significant (Pairwise Wilcoxon $p < 0.01$ with Bonferroni adjustment for both). By 2014, the colonies at Dry Rocks had grown to an average maximum skeletal diameter of 56.38 ± 6.09 cm. The only colony alive at Pickles Reef in 2014 had a maximum diameter of 26.00 cm. Due to the low sample size, comparative statistics could not be performed for 2014 both within and between resamplings. Differences in average maximum skeletal diameter between 2009 and 2010 were statistically significant for all three reefs (Wilcoxon test $p < 0.0001$). Differences in average maximum skeletal diameter between 2010 and 2014 were not statistically significant for Dry Rocks (Wilcoxon test $p = 0.057$).

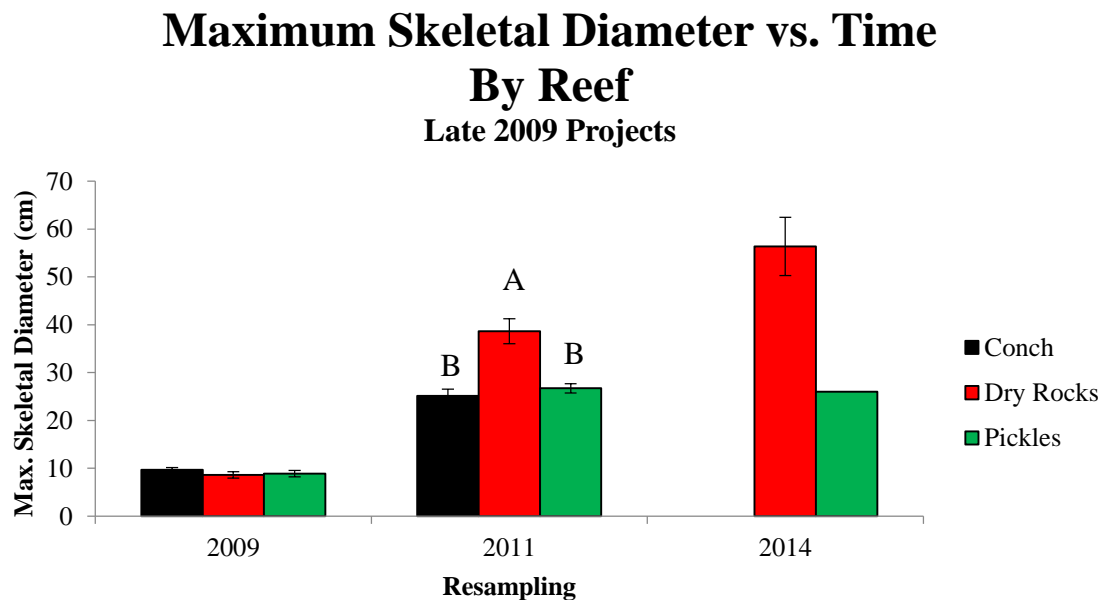


Figure 34. Maximum skeletal diameter by resampling separated by reef for late 2009 projects.

Date Ranges = 2009: 07/13 - 10/29/2009, 2011: 12/10/2010 - 6/15/2011, 2014: 10/01 - 10/29/2014

Totals = Conch - 2009: 48 colonies, 2011: 34; Dry Rocks - 2009: 24, 2011: 21, 2014: 16; Pickles Reef - 2009: 24, 2011: 22, 2014: 1

Maximum Depth: 9m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

When analyzed by genotype, K2 was significantly larger than K3 (Pairwise Wilcoxon $p < 0.0001$ with Bonferroni adjustment), and K1 was larger than either K2 or K3 (Pairwise Wilcoxon $p < 0.05$ and $p < 0.0001$ with Bonferroni adjustment, respectively) at the initial deployment in 2009. In 2009, average maximum skeletal diameter was 11.82 ± 0.52 cm for K1, 9.91 ± 0.35 cm for K2, and 5.95 ± 0.27 cm for K3 (Figure 32). In 2011, the average maximum skeletal diameter had increased to 31.04 ± 1.97 cm for K1, 29.31 ± 2.24 cm for K2, and 27.43 ± 1.85 cm for K3. By 2014, the K1 colonies had an average maximum skeletal diameter of 61.67 ± 9.89 cm, while K2 and K3 colonies recorded 59.50 ± 10.87 cm and 43.75 ± 11.43 cm, respectively. The 2011 and 2014 differences were not statistically significant. However, low sample size by 2014 resulted in high variance.

Differences in average maximum skeletal diameter between 2009 and 2010 were statistically significant for all three reefs (Wilcoxon test $p < 0.0001$). Differences in average maximum skeletal diameter between 2010 and 2014 were statistically significant for K1 and K2 (Wilcoxon test $p < 0.05$). Differences in average maximum skeletal diameter between 2010 and 2014 were not significant for K3 (Wilcoxon test $p = 0.56$).

Maximum Skeletal Diameter vs. Time By Genotype Late 2009 Projects

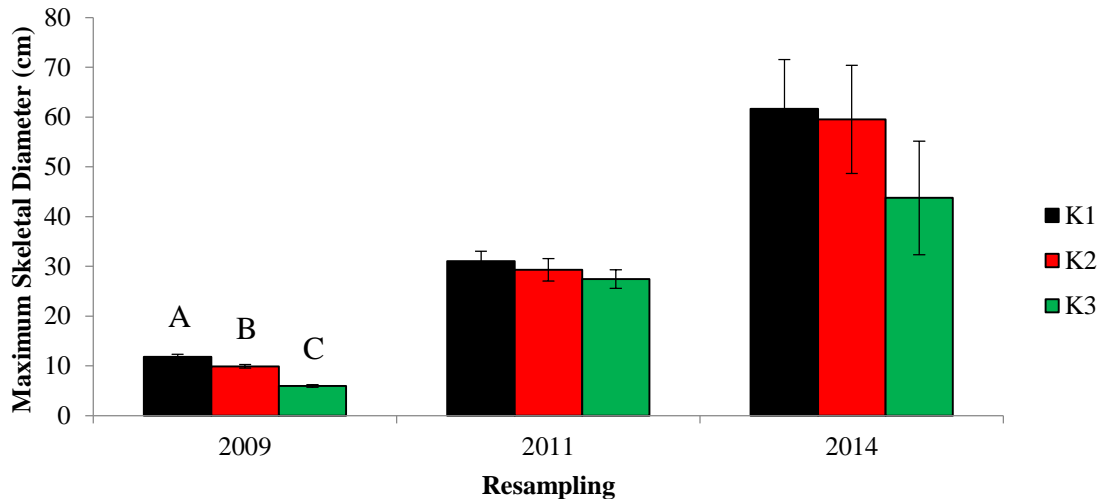


Figure 35. Maximum skeletal diameter by resampling separated by genotype for late 2009 projects.

Date Ranges = 2009: 07/13 - 10/29/2009, 2011: 12/10/2010 - 6/15/2011,
2014: 10/01 - 10/29/2014

Totals = K1 - 2009: 32 colonies, 2011: 26, 2014: 6; K2 – 2009: 32, 2011: 26, 2014: 6;
K3 – 2009: 32, 2011: 25, 2014: 4

Maximum Depth: 9m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

When analyzed by habitat type, a statistical difference was found only in 2011 (Wilcoxon $p < 0.0001$). In 2009, average maximum skeletal diameter was 9.43 ± 0.39 cm for hard-bottom reefs (Conch and Pickles Reefs) and 8.63 ± 0.67 cm for spur-and-groove habitat (Dry Rocks). In 2011, the average maximum skeletal diameter had increased to 25.78 ± 0.93 cm for hard-bottom sites and 38.64 ± 2.60 cm for spur-and-groove (Figure 33). By 2014, only one colony remained on hard-bottom substrate (maximum skeletal diameter of 26.00 cm). The average maximum skeletal diameter for spur-and-groove colonies was of 56.38 ± 6.09 cm. Due to the low sample size, comparative statistics could not be performed for 2014 both within and between resamplings. Differences in average maximum skeletal diameter between 2009 and 2010 were statistically significant for both habitat types (Wilcoxon test $p \ll 0.0001$).

Maximum Skeletal Diameter vs. Time By Habitat Type Late 2009 Projects

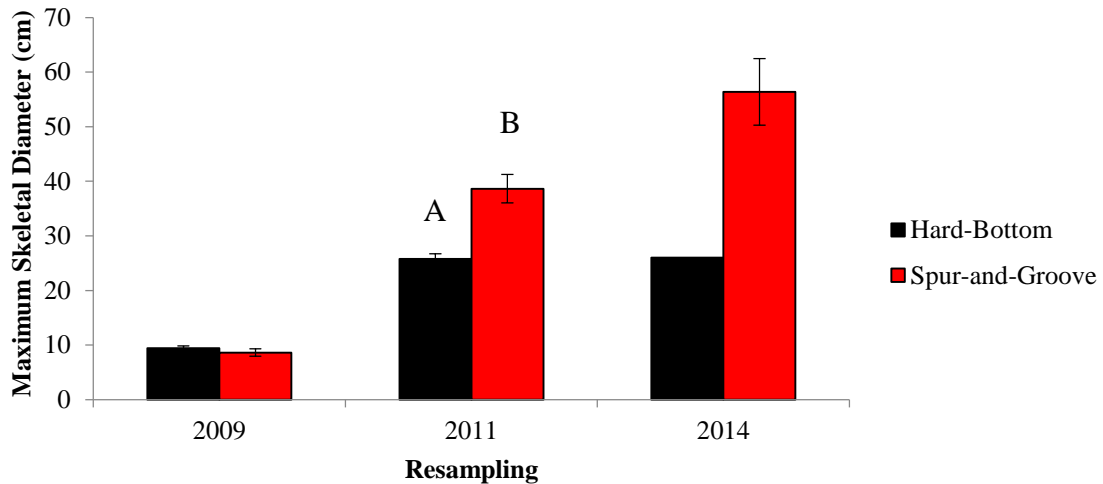


Figure 36. Maximum skeletal diameter by resampling separated by habitat type for late 2009 projects.

Date Ranges = 2009: 07/13 - 10/29/2009, 2011: 12/10/2010 - 6/15/2011, 2014: 10/01 - 10/29/2014

Totals = Hard-bottom - 2009: 72 colonies, 2011: 56, 2014: 1;

Spur-and-Groove – 2009: 24, 2011: 21, 2014: 16

Maximum Depth: 9m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

For percent survival and percent live tissue cover in 2011, differences between the Conch Reef projects and the other sites were detected (Figures 34 and 35). Survival at CS1 was $54.17 \pm 10.39\%$, compared to $87.50 \pm 6.90\%$ for CS2, 100% for DR2 and $91.67 \pm 5.76\%$ for PK2. The differences between CS1 and DR2 along with PK2 were statistically significant (Pairwise Wilcoxon $p < 0.01$ and $p < 0.05$ with Bonferroni adjustment, respectively). CS1 had significantly less live tissue coverage than the other three projects (Pairwise Wilcoxon $p < 0.01$ with Bonferroni adjustment for all). CS1 averaged $34.29 \pm 9.01\%$ live tissue coverage compared to $81.08 \pm 7.43\%$ for CS2, $99.90 \pm 0.07\%$ for DR2, and $87.29 \pm 6.10\%$ for PK2. In 2014, the differences between Dry Rocks and Pickles Reef for both percent survival and live tissue coverage was statistically significant (Wilcoxon $p < 0.0001$ for both). Survival at Dry Rocks was

$66.67 \pm 9.83\%$ with $35.00 \pm 7.85\%$ live tissue coverage. Pickles Reef had only one live colony remaining (3.57% survival) with <1% live tissue coverage.

Differences in percent survival and percent live tissue between 2009 and 2011 were statistically significant for CS1 (Wilcoxon test $p < 0.001$). For CS2, the difference in percent survival between 2009 and 2011 was not significant (Wilcoxon test $p = 0.081$) but was significant for percent live tissue (Wilcoxon test $p < 0.01$). All of the colonies transplanted to Dry Rocks survived with almost 100% tissue coverage to 2011. The differences in percent survival and percent live tissue between 2011 and 2014 were statistically significant (Wilcoxon test $p < 0.01$). At PK2, there was no statistically significant difference in percent survival between 2009 and 2011 (Wilcoxon $p = 0.48$). There difference in percent live tissue was significant (Wilcoxon $p < 0.05$). The differences between 2011 and 2014 for both percent survival and percent live tissue were significant (Wilcoxon $p < 0.0001$).

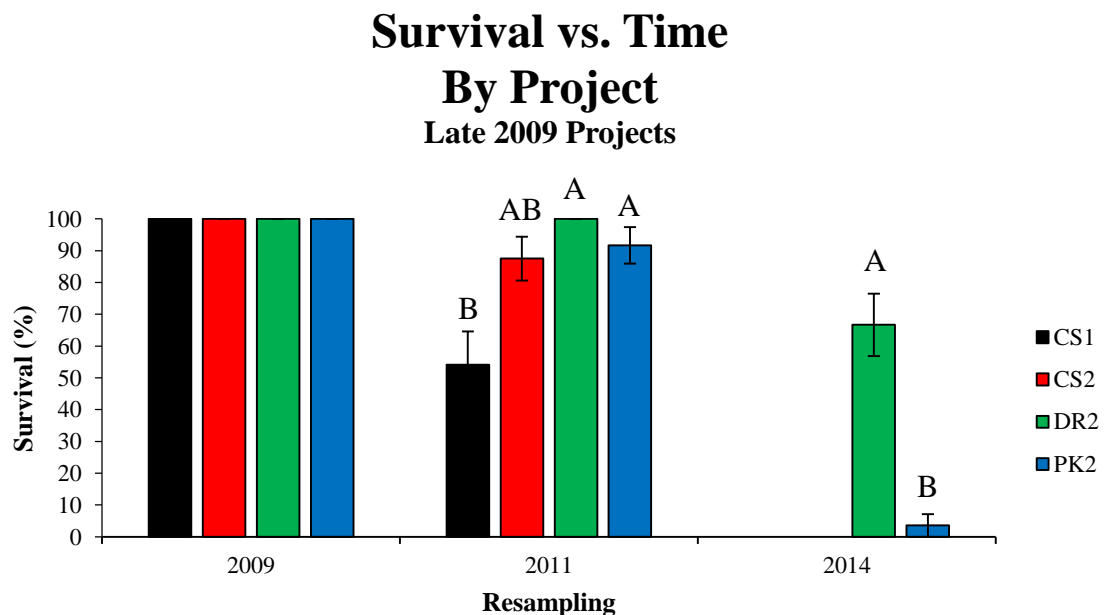


Figure 37. Colony survivorship by resampling separated by project for late 2009 projects. ((# alive/total)*100)

Date Ranges = 2009: 07/13 - 10/29/2009, 2011: 12/10/2010 - 6/15/2011, 2014: 10/01 - 10/29/2014

Totals = Conch #1 - 2009: 24 colonies, 2011: 24, Conch #2: 2009: 24, 2011: 24, Dry Rocks - 2009: 24, 2011: 21, 2014: 24; Pickles - 2009: 24, 2011: 24, 2014: 1
Maximum Depth: 9 m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

Percent Live Tissue vs. Time By Project

Late 2009 Projects

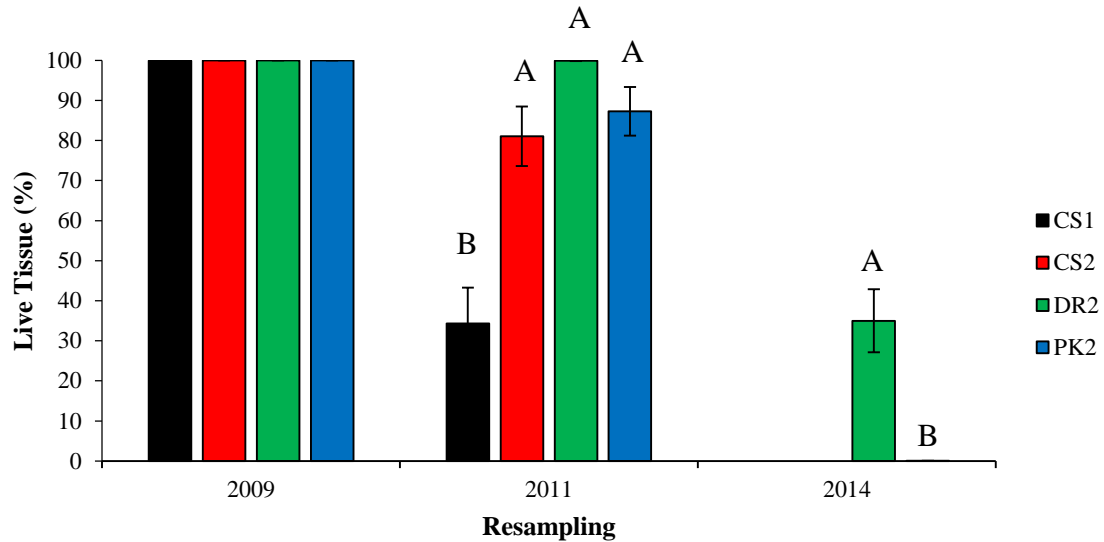


Figure 38. Percent live tissue by resampling separated by project for late 2009 projects.

Date Ranges = 2009: 07/13 - 10/29/2009, 2011: 12/10/2010 - 6/15/2011,

2014: 10/01 - 10/29/2014

Totals = Conch #1 - 2009: 24 colonies, 2011: 24, Conch #2: 2009: 24, 2011: 24,

Dry Rocks - 2009: 24, 2011: 21, 2014: 24; Pickles - 2009: 24, 2011: 24, 2014: 1

Maximum Depth: 9m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

No statistically significant differences were observed for either percent survival or live tissue coverage based on genotype (Figures 36 and 37). In 2011, survival of K1 colonies was $83.87 \pm 6.72\%$ with $77.52 \pm 6.91\%$ live tissue coverage. K2 transplants had the same percent survival but a slightly lower average percent live tissue coverage ($73.71 \pm 7.64\%$). K3 colonies averaged $80.65 \pm 7.21\%$ survival with $73.35 \pm 7.62\%$ live tissue coverage. By 2014, average percent survival had declined slightly to $75.00 \pm 16.37\%$ for both K1 and K2, and $50.00 \pm 18.90\%$ for K3. Average percent live tissue coverage displayed a slightly larger decline as K1 fell to $46.38 \pm 14.33\%$, K2 dropped to $40.75 \pm 13.26\%$, and K3 declined to $17.88 \pm 12.71\%$.

Percent survival was not statistically significantly different for K1 or K2 between either 2009 and 2011 or 2011 and 2014 (both Wilcoxon $p = 0.059$ and $p = 1.00$,

respectively). However, the difference between 2009 and 2014 was significant (Wilcoxon $p < 0.05$). The differences in percent live tissue between 2009 and 2011 were significant (Wilcoxon $p < 0.01$), but not significant between 2011 and 2014 (Wilcoxon $p = 0.054$ (K1) and $p = 0.080$ (K2)). The difference in percent survival between 2009 and 2011 was statistically significant for K3 (Wilcoxon test $p < 0.05$), but was not significant between 2011 and 2014 (Wilcoxon $p = 0.25$). For percent live tissue, differences between all three time-steps were significant (Wilcoxon $p < 0.05$).

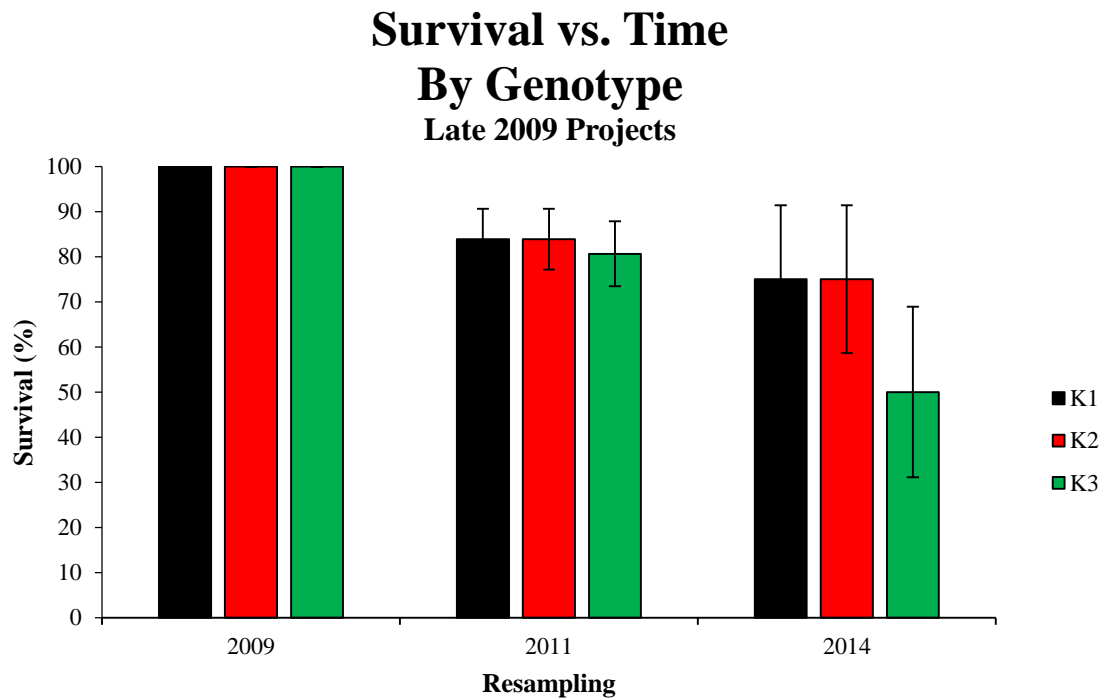


Figure 39. Colony survivorship by resampling separated by genotype for late 2009 projects.

((# alive/total)*100)

Date Ranges = 2009: 07/13 - 10/29/2009, 2011: 12/10/2010 - 6/15/2011,
2014: 10/01 - 10/29/2014

Totals = K1 - 2009: 32 colonies, 2011: 31, 2014: 8; K2: 2009: 32, 2011: 31, 2014: 8,
K3 - 2009: 32, 2011: 31, 2014: 8

Maximum Depth: 9m

Error bars indicate standard error.

Live Tissue vs. Time By Genotype Late 2009 Projects

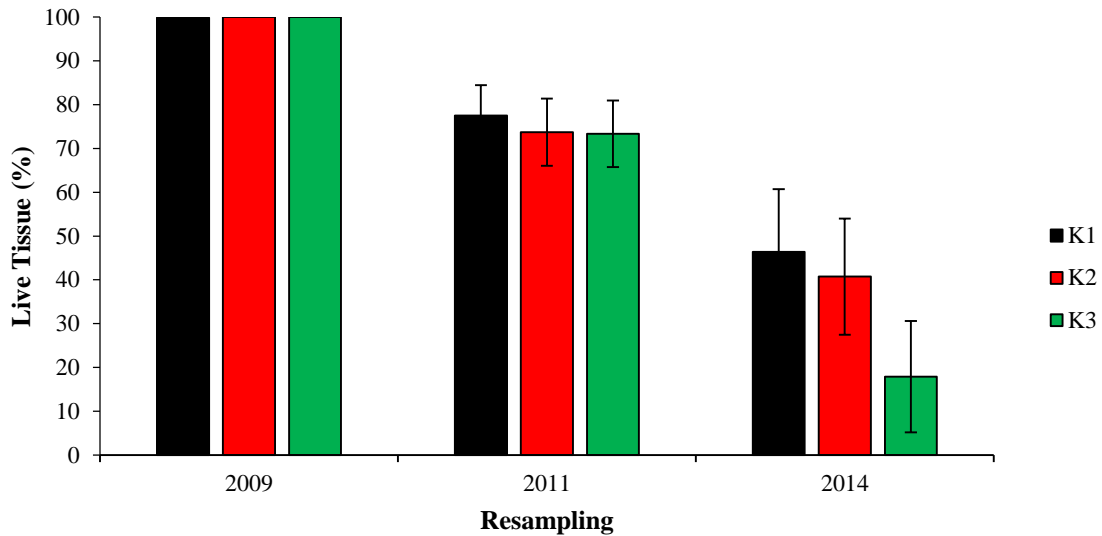


Figure 40. Percent live tissue by resampling separated by genotype for late 2009 projects. Date Ranges = 2009: 07/13 - 10/29/2009, 2011: 12/10/2010 - 6/15/2011, 2014: 10/01 - 10/29/2014
Totals = K1 - 2009: 32 colonies, 2011: 31, 2014: 8; K2: 2009: 32, 2011: 31, 2014: 8, K3 - 2009: 32, 2011: 31, 2014: 8
Maximum Depth: 9m
Error bars indicate standard error.

When analyzed by habitat type, significant differences in both percent survival and live tissue coverage were detected (Figures 38 and 39). Hard-bottom habitats (Conch and Pickles Reefs) had an average percent survival of $77.78 \pm 4.93\%$ in 2011 compared to 100% at the spur-and-groove site (Dry Rocks, Wilcoxon $p < 0.05$). Percent live tissue coverage averaged $67.56 \pm 5.16\%$ for hard-bottom sites and $99.90 \pm 0.07\%$ for spur-and-groove (Wilcoxon $p < 0.01$). In 2014, average percent survival on hard-bottom structures declined to $3.57 \pm 3.57\%$ versus $66.67 \pm 9.83\%$ on spur-and-groove substrate (Wilcoxon $p < 0.0001$). Average percent live tissue had fallen to $0.04 \pm 0.04\%$ for hard-bottom compared to $35.00 \pm 7.85\%$ for spur-and-groove (Wilcoxon $p < 0.0001$). Differences in percent survival and percent live tissue all three time-steps were statistically significant for hard-bottom sites (Wilcoxon test $p < 0.0001$). The spur-and-groove site had 100%

and almost 100% live tissue in 2011. The differences in percent survival and percent live tissue between 2011 and 2014 were statistically significant (Wilcoxon test $p < 0.01$).

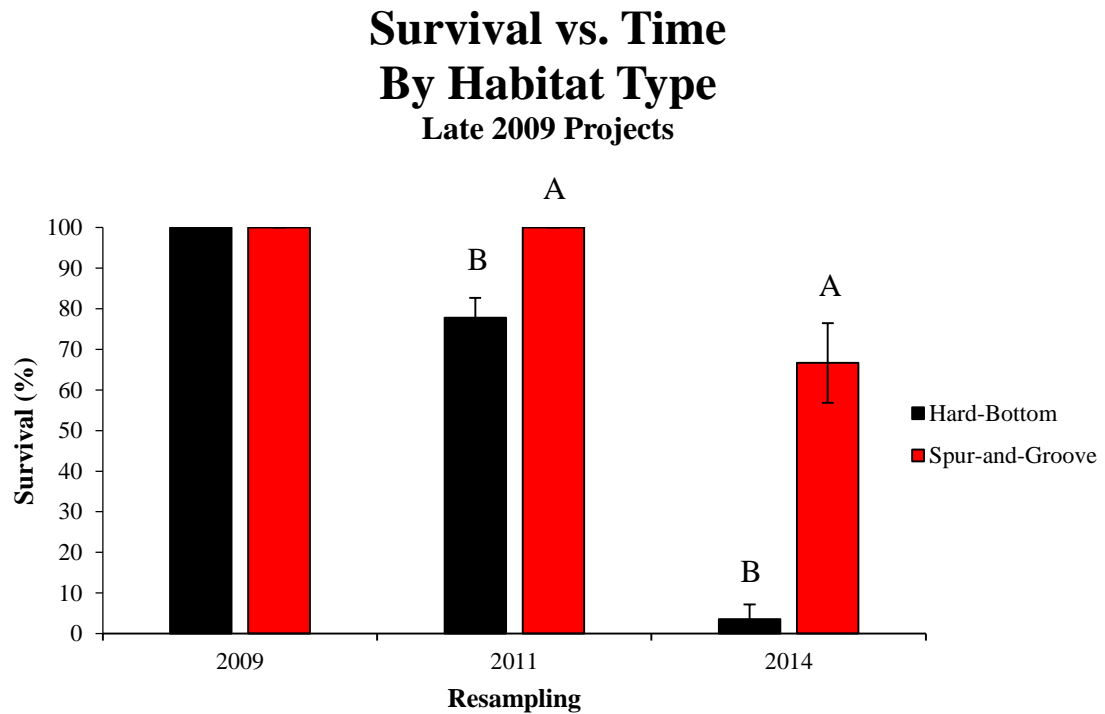


Figure 41. Colony survivorship by resampling separated by habitat type for late 2009 projects.

((# alive/total)*100)

Date Ranges = 2009: 07/13 - 10/29/2009, 2011: 12/10/2010 - 6/15/2011,
2014: 10/01 - 10/29/2014

Totals = Hard-bottom - 2009: 72 colonies, 2011: 72, 2014: 28;

Spur-and-groove: 2009: 24, 2011: 21, 2014: 24

Maximum Depth: 9m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

Percent Live Tissue vs. Time By Habitat Type Late 2009 Projects

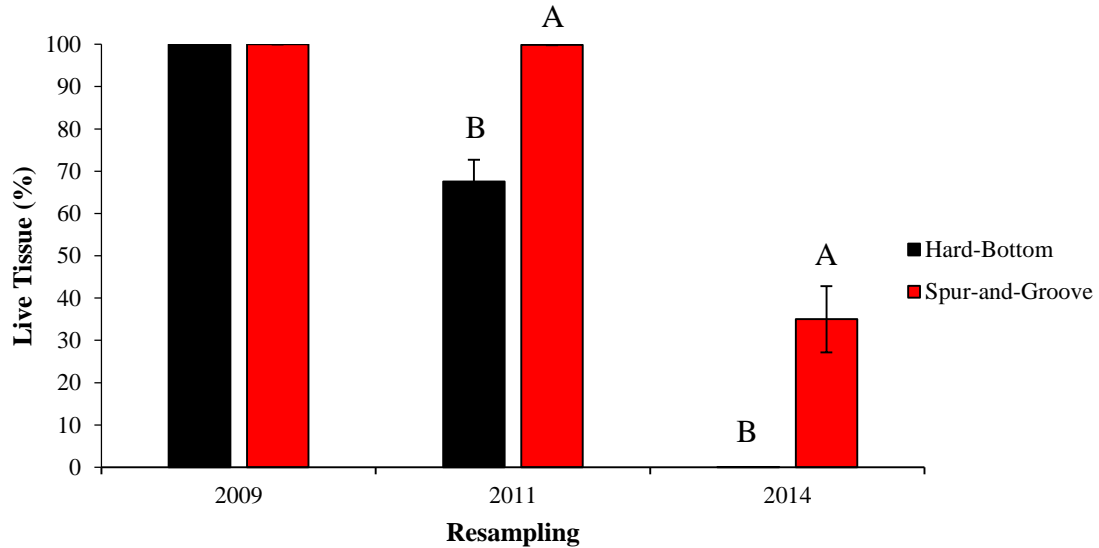


Figure 42. Percent live tissue by resampling separated by habitat type for late 2009 projects.

Date Ranges = 2009: 07/13 - 10/29/2009, 2011: 12/10/2010 - 6/15/2011, 2014: 10/01 - 10/29/2014

Totals = Hard-bottom - 2009: 72 colonies, 2011: 72, 2014: 28;

Spur-and-groove: 2009: 24, 2011: 21, 2014: 24

Maximum Depth: 9m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

2010 PROJECTS

Two projects were conducted in 2010 – one at French Reef and one at Molasses Trench. Twenty four corals each were deployed in the triangular configuration using eight colonies each from the K1, K2, and K3 genotypes. Rubber genotype tags were epoxied to the base of the colonies. The French Reef project (project ID “FR2”) was started on May 27, 2010 on spur at a maximum depth of 9m under French Ball #7. The Molasses Trench project (project ID “MT2”) began on July 30, 2010 at a maximum depth of 8m on a patch reef within the Molasses Trench, 1.72km NNW of the main reef. CRF conducted one monitoring visit to both sites in 2011. *In situ* monitoring was conducted at FR2 on September 2, 2014. No *in situ* visits were conducted at MT2 due to logistical difficulties.

The average (\pm SE) initial maximum skeletal diameter for the 48 colonies deployed was 9.04 ± 0.34 cm. By 2011, the average maximum skeletal diameter had increased to 20.67 ± 1.07 cm (Figure 40). The 2014 in situ measurements from the two remaining live colonies on French Reef had increased to an average diameter of 55.00 ± 19.00 cm. Difference in average maximum skeletal diameter between 2009 and 2010 was statistically significant (Wilcoxon test $p \ll 0.0001$). Low sample size in 2014 prevented statistical comparison.

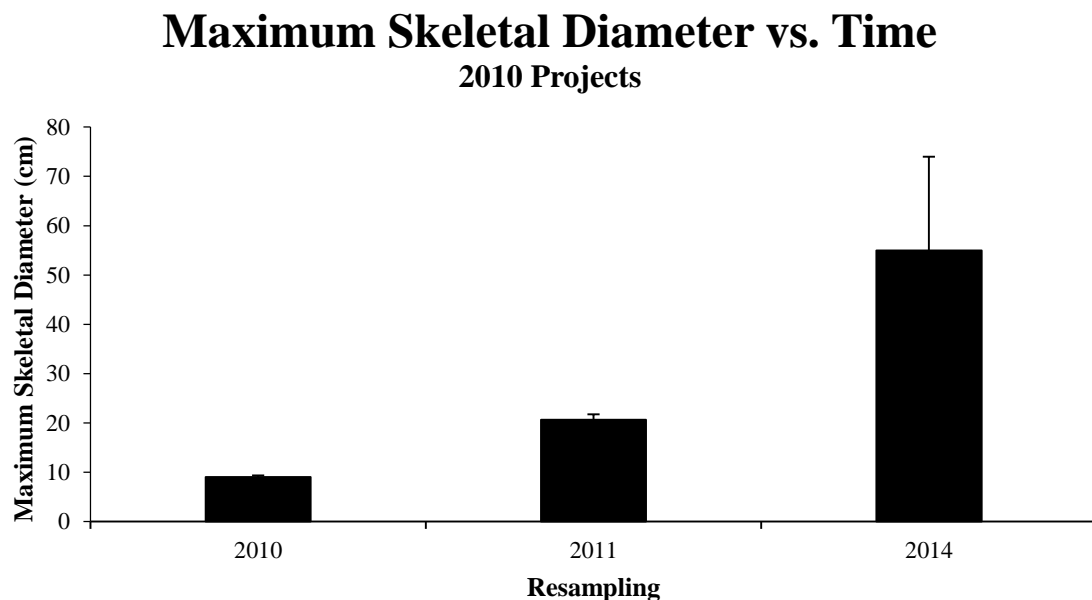


Figure 43. Maximum skeletal diameter by resampling for 2010 projects. Data averaged across both reefs (Molasses Trench and French Reef). Date Ranges = 2010: 05/27 - 07/30/2010, 2011: 02/09 - 07/12/2011, 2014: 09/02/2014 Totals = 2010: 48 colonies, 2011: 34, 2014: 2 Maximum Depth: 9m Error bars indicate standard error.

When analyzed by reef, no statistical differences were identified at any time. In 2010, average maximum skeletal diameter was 9.44 ± 0.44 cm for Molasses Trench, and 8.57 ± 0.53 cm for French Reef (Figure 41). In 2010, the average maximum skeletal diameter had increased to 23.36 ± 1.91 cm for Molasses Trench and 18.79 ± 1.08 cm for French Reef. Only two colonies remained alive in 2014 at French Reef, with an average maximum skeletal diameter of 55.00 ± 19.00 cm. A comparison based on habitat type is identical to the above comparison, where “MT2” can be replaced with “patch reef” and

“FR2” with “spur-and-groove” (Figure 46). Differences in average maximum skeletal diameter between 2009 and 2010 were statistically significant for both reefs (Wilcoxon test $p < 0.0001$). Low sample size in 2014 prevented statistical comparison.

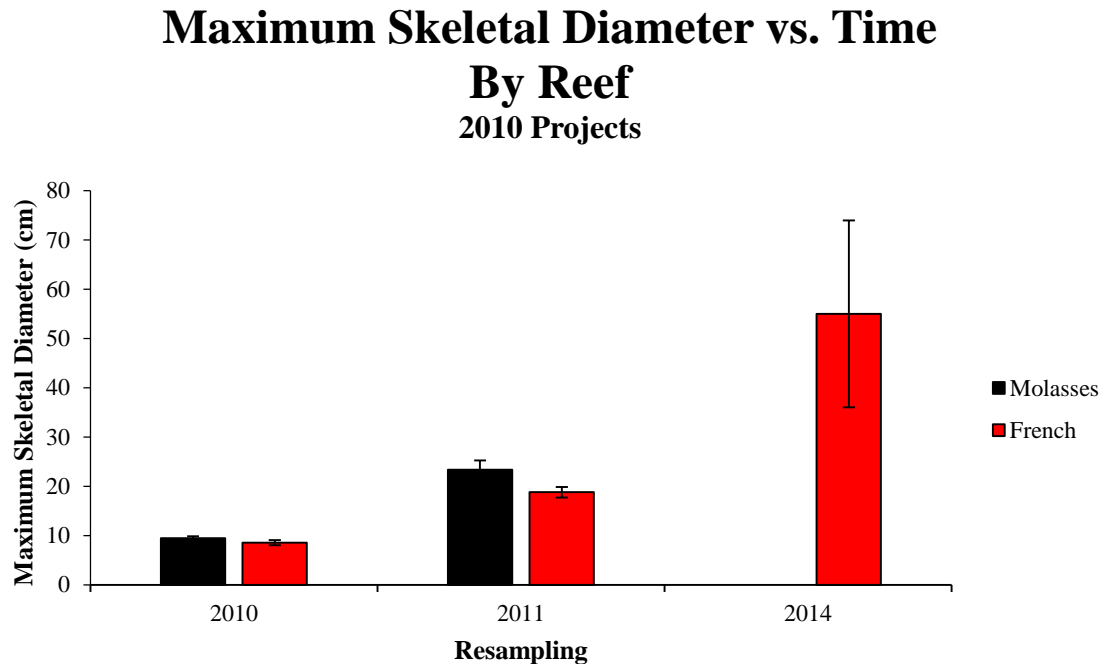


Figure 44. Maximum skeletal diameter by resampling separated by reef for 2010 projects. Date Ranges = 2010: 05/27 - 07/30/2010, 2011: 02/09 - 07/12/2011, 2014: 09/02/2014
Totals = Molasses Trench - 2010: 24 colonies, 2011: 14;
French Reef - 2010: 24, 2011: 20, 2014: 2
Maximum Depth: 9m
Error bars indicate standard error.

When analyzed by genotype, K3 was initially smaller than both K1 and K2 but no trend was identified in the 2011 analysis (Pairwise Wilcoxon $p < 0.05$ and $p < 0.001$ with Bonferroni adjustment, respectively). In 2010, K1 colonies had an average maximum skeletal diameter of 9.44 ± 0.50 cm, while K2 and K3 recorded 10.51 ± 0.47 cm and 7.16 ± 0.48 cm, respectively (Figure 42). After one year on the reef, the average maximum skeletal diameter of the colonies increased to 18.51 ± 1.85 cm for K1, 22.53 ± 1.42 cm for K2, and 20.80 ± 2.23 cm for K3. In 2014, one K1 colony had grown to 36.00 cm and one K2 colony reached 74.00 cm. Differences in average maximum skeletal diameter between 2009 and 2010 were statistically significant for all three genotypes (Wilcoxon test $p < 0.001$). Low sample size in 2014 prevented statistical comparison.

Maximum Skeletal Diameter vs. Time By Genotype

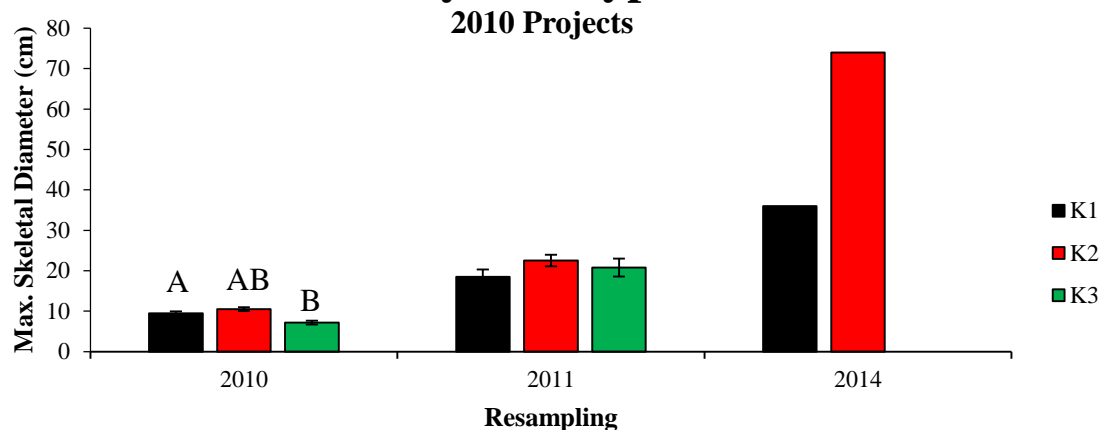


Figure 45. Maximum skeletal diameter by resampling separated by genotype for 2010 projects.

Date Ranges = 2010: 05/27 - 07/30/2010, 2011: 02/09 - 07/12/2011, 2014: 09/02/2014

Totals = K1 - 2010: 16 colonies, 2011: 11, 2014: 1; K2 - 2010: 16, 2011: 12, 2014: 1;

K3 - 2010: 16, 2011: 11

Maximum Depth: 9m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

2014 analysis is for FR2 only.

Maximum Skeletal Diameter vs. Time By Habitat Type

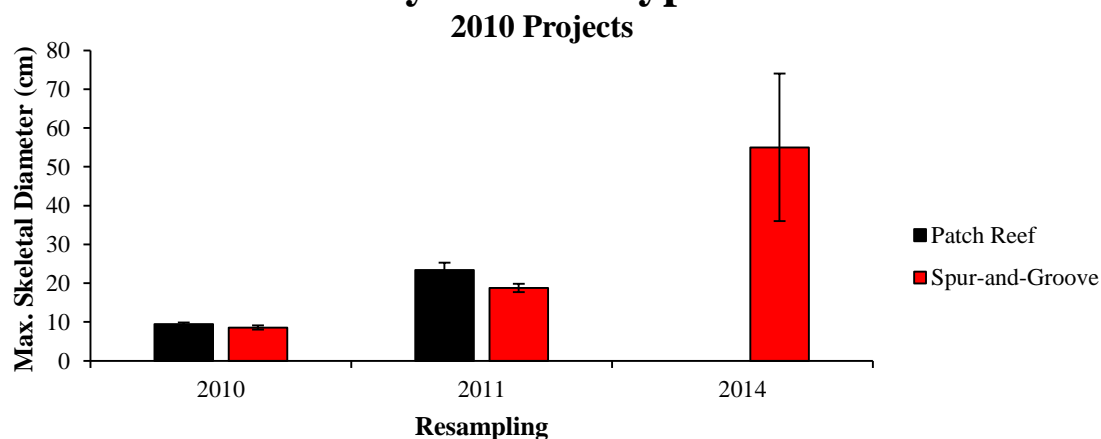


Figure 46. Maximum skeletal diameter by resampling separated by habitat type for 2010 projects.

Date Ranges = 2010: 05/27 - 07/30/2010, 2011: 02/09 - 07/12/2011, 2014: 09/02/2014

Totals = Molasses Trench - 2010: 24 colonies, 2011: 14;

French Reef - 2010: 24, 2011: 20, 2014: 2

Maximum Depth: 9m

Error bars indicate standard error.

In 2011, MT2 recorded $58.33 \pm 10.28\%$ survival with $55.54 \pm 10.00\%$ live tissue coverage, while FR2 had $83.33 \pm 7.77\%$ survival with $81.25 \pm 7.86\%$ live tissue coverage (Figures 43 and 44). Only the difference in average percent live tissue coverage was statistically significant (Wilcoxon $p < 0.05$). As in the skeletal diameter analysis, comparison based on habitat type for survival or condition is identical to the above comparison, where “MT1” and “FR1” with “patch reef” and “spur-and-groove”, respectively (Figures 51 and 52). Differences in percent survival and percent live tissue between 2010 and 2011 were not statistically significant for French Reef (Wilcoxon test $p = 0.12$ and $p = 0.22$, respectively), but were significant between 2011 and 2014 (Wilcoxon test $p < 0.0001$). Differences in percent survival and percent live tissue were significant for Molasses Trench between 2010 and 2011 (Wilcoxon $p < 0.001$).

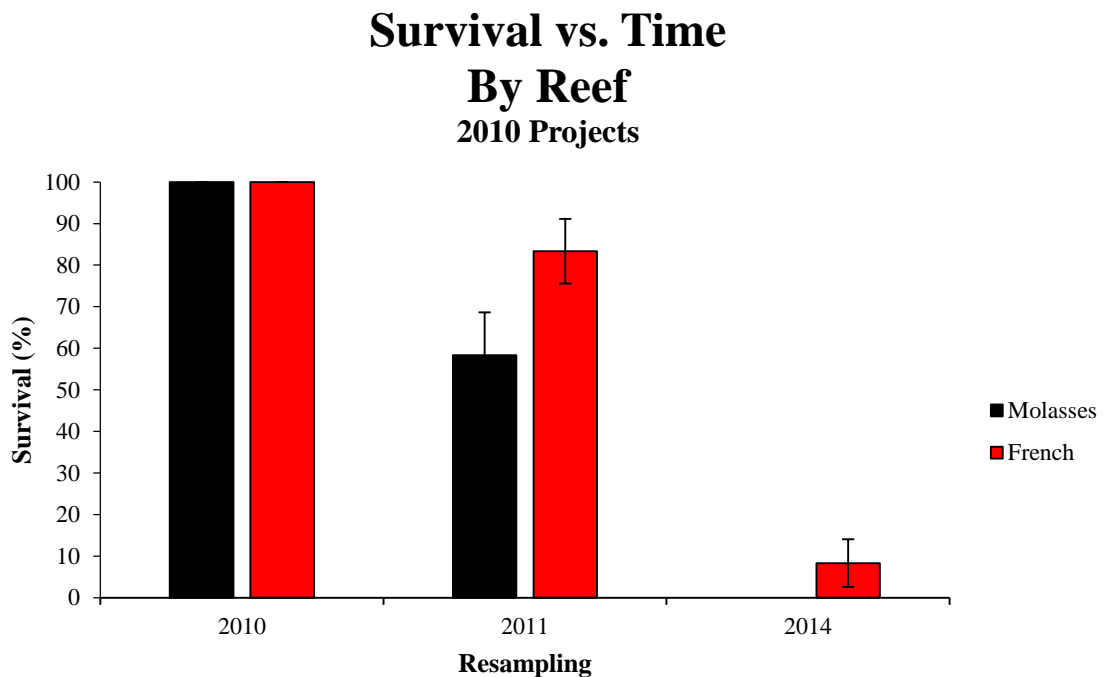


Figure 47. Colony survivorship by resampling separated by reef for 2010 projects.
((# alive/total)*100)

Date Ranges = 2010: 05/27 - 07/30/2010, 2011: 02/09 - 07/12/2011, 2014: 09/02/2014

Totals = Molasses Trench - 2010: 24 colonies, 2011: 24;

French Reef - 2010: 24, 2011: 24, 2014: 24

Maximum Depth: 9m

Error bars indicate standard error.

Percent Live Tissue vs. Time By Reef 2010 Projects

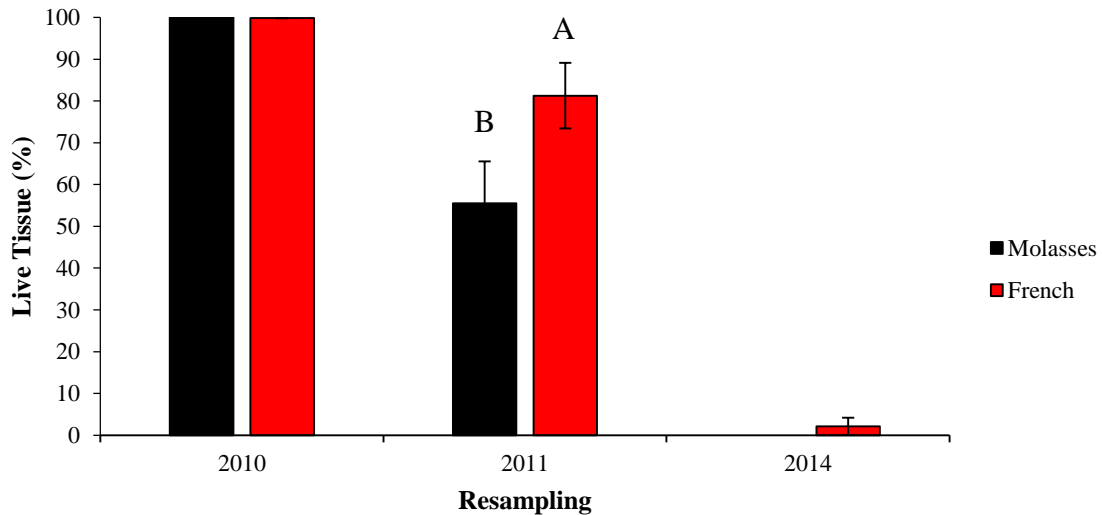


Figure 48. Percent live tissue by resampling separated by reef for 2010 projects.
 Date Ranges = 2010: 05/27 - 07/30/2010, 2011: 02/09 - 07/12/2011, 2014: 09/02/2014
 Totals = Molasses Trench - 2010: 24 colonies, 2011: 24;
 French Reef - 2010: 24, 2011: 24, 2014: 24
 Maximum Depth: 9m
 Error bars indicate standard error. Letters indicate statistical significance within sampling.

There were no statistically significant differences when corals were analyzed by genotype for either percent survival or percent live tissue coverage at any time. In 2011, $68.75 \pm 11.97\%$ of the K1 colonies, $75.00 \pm 11.18\%$ of the K2 colonies, and $68.75 \pm 11.97\%$ of the K3 colonies survived (Figure 45). Average percent live tissue coverage was $67.69 \pm 11.80\%$ for K1, $71.88 \pm 11.15\%$ for K2, and $65.63 \pm 11.83\%$ for K3 (Figure 46). By the 2014 *in situ* resampling at French Reef, only two colonies remained alive – one each from K1 and K2. The K1 colony had $<1\%$ live tissue while the K2 colony had 5%.

Difference in percent survival between 2010 and 2011 was not statistically significant for all three genotypes (Wilcoxon test $p = 0.054$ (K1 and K3), $p = 0.11$ (K2)), but was significant between 2011 and 2014 (Wilcoxon test $p < 0.05$). Percent live tissue for both K1 and K3 was significantly different between all three time-steps (Wilcoxon

test $p < 0.05$). Difference in percent live tissue was not significant for K2 between 2010 and 2011 (Wilcoxon $p = 0.055$), but was significant between 2011 and 2014 (Wilcoxon $p < 0.01$).

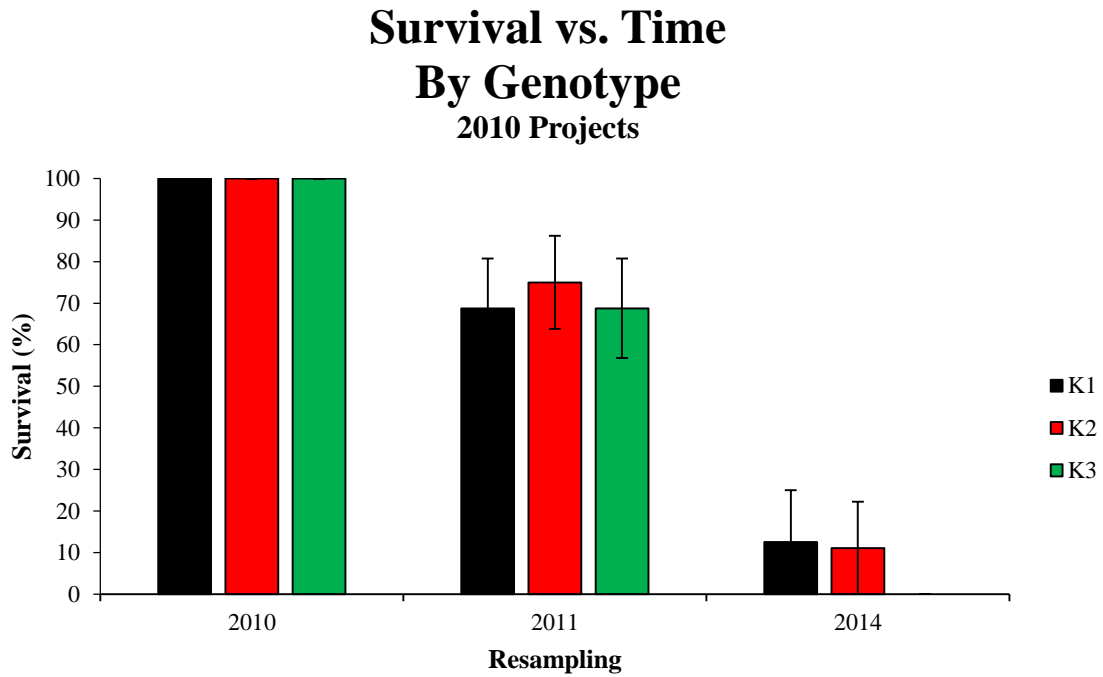


Figure 49. Colony survivorship by resampling separated by genotype for 2010 projects. Date Ranges = 2010: 05/27 - 07/30/2010, 2011: 02/09 - 07/12/2011, 2014: 09/02/2014
Totals = K1 - 2010: 16 colonies, 2011: 16, 2014: 8; K2 - 2010: 16, 2011: 16, 2014: 8; K3 - 2010: 16, 2011: 16, 2014: 7
Maximum Depth: 9m
Error bars indicate standard error.
2014 analysis is for FR2 only.

Percent Live Tissue vs. Time By Genotype

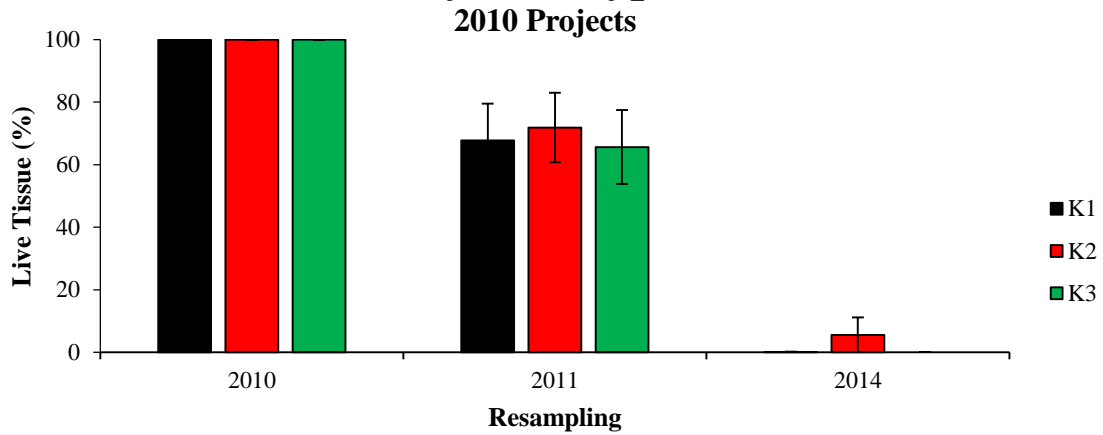


Figure 50. Percent live tissue by resampling separated by genotype for 2010 projects.
 Date Ranges = 2010: 05/27 - 07/30/2010, 2011: 02/09 - 07/12/2011, 2014: 09/02/2014
 Totals = K1 - 2010: 16 colonies, 2011: 16, 2014: 8; K2 - 2010: 16, 2011: 16, 2014: 8;
 K3 - 2010: 16, 2011: 16, 2014: 7
 Maximum Depth: 9m
 Error bars indicate standard error.
 2014 analysis is for FR2 only.

Survival vs. Time By Habitat Type

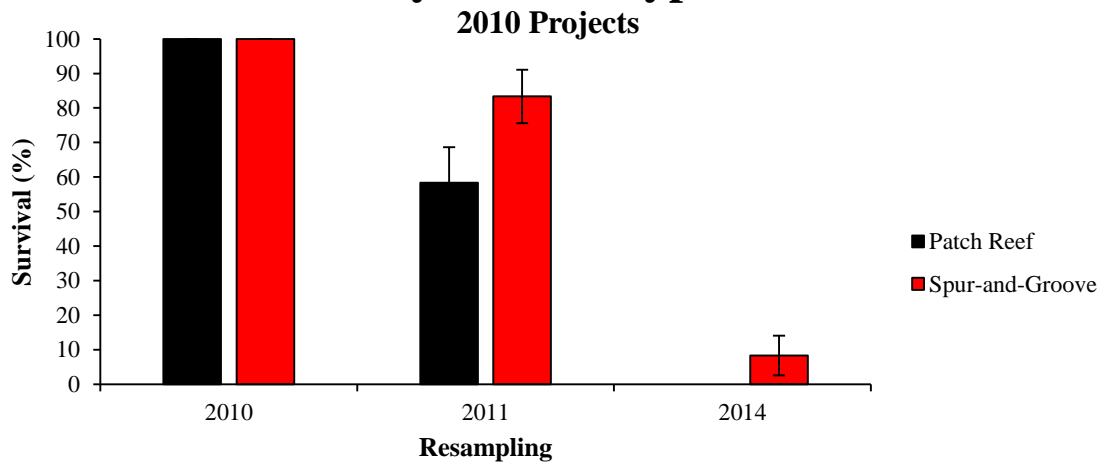


Figure 51. Colony survivorship by resampling separated by habitat type for 2010 projects.
 ((# alive/total)*100)
 Date Ranges = 2010: 05/27 - 07/30/2010, 2011: 02/09 - 07/12/2011, 2014: 09/02/2014
 Totals = Molasses Trench - 2010: 24 colonies, 2011: 24;
 French Reef - 2010: 24, 2011: 24, 2014: 24
 Maximum Depth: 9 m
 Error bars indicate standard error.

Percent Live Tissue vs. Time By Habitat Type 2010 Projects

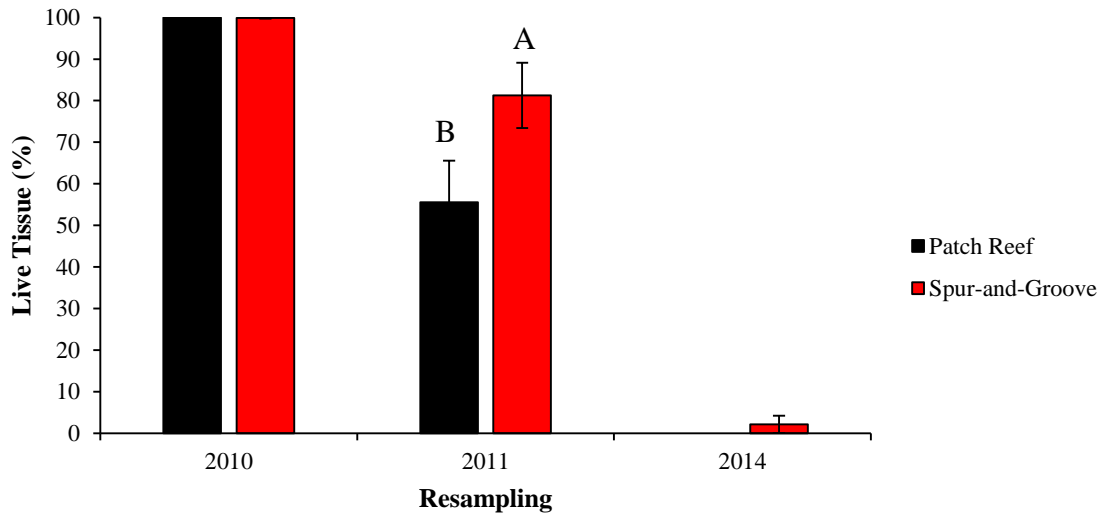


Figure 52. Percent live tissue by resampling separated by habitat type for 2010 projects. Date Ranges = 2010: 05/27 - 07/30/2010, 2011: 02/09 - 07/12/2011, 2014: 09/02/2014
Totals = Molasses Trench - 2010: 24 colonies, 2011: 24;
French Reef - 2010: 24, 2011: 24, 2014: 24
Maximum Depth: 9 m
Error bars indicate standard error. Letters indicate statistical significance within sampling.

2012 PROJECTS

Transplanting in 2012 was funded through a grant as part of the American Restoration and Reinvestment Act. Four reefs were selected as part of the project: Pickles Reef, Conch Reef, Molasses Reef, and Dry Rocks. Each site received 400 *A. cervicornis* colonies. However, they were not all recorded in the Coral Restoration Foundation photographic record due to logistical challenges coordinating man-power for the transplanting, in-water time, and documentation. From the CRF photographs, 304 individuals were initially recorded at Pickles Reef (project ID “PKARRA”), 278 colonies from Conch Reef (project ID “CNARRA”), 312 colonies from Molasses Reef (project ID “MLARRA”), and 310 individuals from Dry Rocks (project ID “DRARRA”). Long plastic tags indicating genotype and an individual identifier were epoxied to the substrate for 50 colonies from each reef. Only these 50 colonies were photographed during

maintenance visits. The remaining colonies at each reef had small plastic tags indicating their genotype attached to a branch with wire. A total of 14 genotypes were used, 10 of which were utilized at all four sites. The colonies were attached to the reef in loose clusters based on available space. All of the colonies observed during the 2014 *in situ* resampling were recorded.

PKARRA was started on April 24, 2012 using 11 genotypes at a depth of 6m on a hard-bottom structure between Pickles Balls #1 and #2. CNARRA was transplanted on May 12, 2012 at 8m depth onto a hard-bottom structure between Conch Balls #2 and #3 using 13 genotypes. MLARRA received corals from 10 genotypes on May 22, 2012 at a depth of 7m on a spur-and-groove under Molasses Ball #8. Colonies from 11 genotypes were transplanted to DRARRA on June 5, 2012 at a depth of 8m onto spur-and-groove under Dry Rocks Balls #4 and #5.

Several maintenance visits were included in the photographic record provided by the Coral Restoration Foundation. Pickles Reef was revisited four times on July 9, 2012, September 8, 2012, May 10, 2013, and October 5, 2013. Conch Reef was revisited three times on June 15, 2012, September 8, 2012, and September 12, 2013. Two visits were conducted at Dry Rocks on July 16, 2012, and September 25, 2012. Monitoring on Molasses Reef was performed on July 16, 2012.

The average (\pm SE) initial maximum skeletal diameter for the 1,204 colonies recorded in the initial photos was 16.09 ± 0.17 cm. By 2014, the average maximum skeletal diameter had grown to 30.19 ± 0.62 cm (Figure 47), an increase of 14.10 cm. Difference in average maximum skeletal diameter between 2012 and 2014 was statistically significant (Wilcoxon test $p < 0.0001$).

Maximum Skeletal Diameter vs. Time 2012 Projects

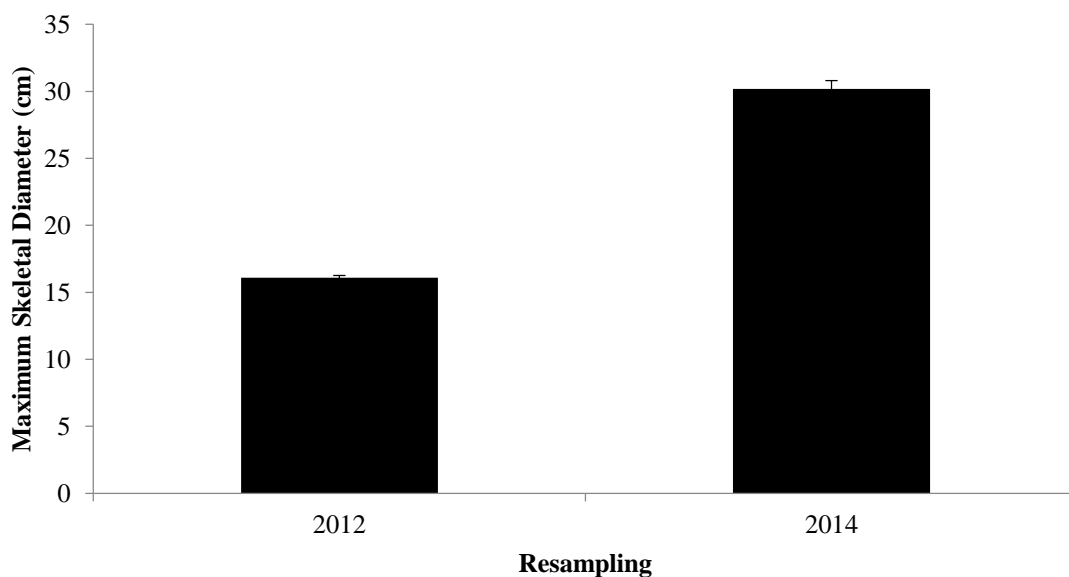


Figure 53. Maximum skeletal diameter by resampling for 2012 projects. Data averaged across all four reefs (Molasses Reef, Pickles Reef, Conch Reef, and Dry Rocks).
 Date Ranges = 2012: 4/24 - 6/6/2012, 2014: 10/29/2014 - 1/22/2015
 Totals = 2012: 1,204 colonies, 2014: 549
 Maximum Depth: 9m
 Error bars indicate standard error.

When analyzed by reef, colonies at Molasses Reef were significantly larger at the initial transplanting compared to the other three sites (pairwise Wilcoxon $p < 0.0001$). In 2012, average maximum skeletal diameter was 19.57 ± 0.34 cm for Molasses Reef, and 14.86 ± 0.24 cm for Conch Reef, 15.19 ± 0.32 cm for Dry Rocks, and 15.04 ± 0.28 cm for Pickles Reef (Figure 48). In 2014, the average maximum skeletal diameter had increased to 32.75 ± 1.46 cm for Molasses Reef, 25.97 ± 1.94 cm for Conch Reef, 28.94 ± 0.97 cm for Pickles Reef, and 30.99 ± 0.96 cm for Dry Rocks. There were no statistical differences in maximum skeletal diameter among reefs in 2014. Differences in average maximum skeletal diameter between 2012 and 2014 were statistically significant for all four reefs (Wilcoxon test $p < 0.0001$).

Maximum Skeletal Diameter vs. Time by Reef 2012 Projects

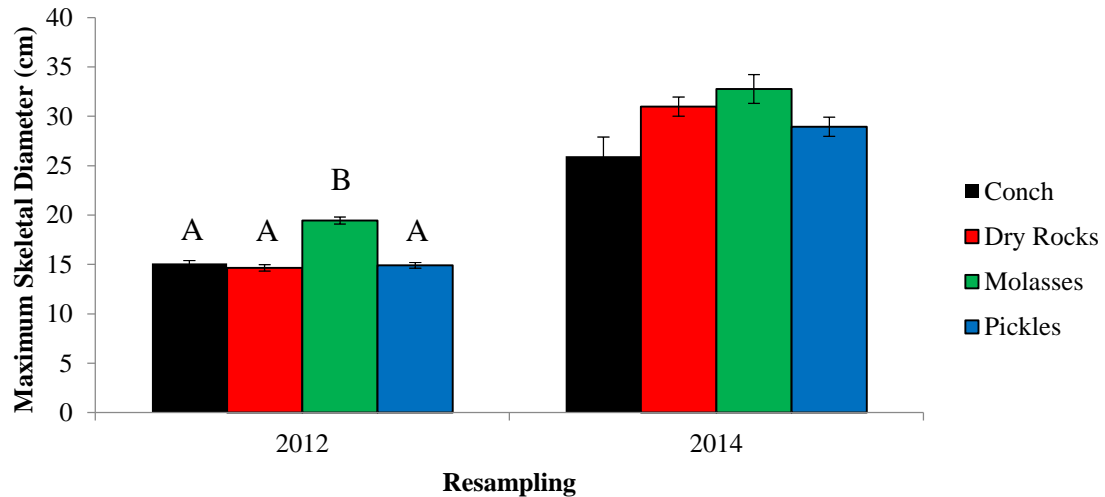


Figure 54. Maximum skeletal diameter by resampling separated by reef for 2012 projects. Date Ranges = 2012: 4/24 - 6/6/2012, 2014: 10/29/2014 - 1/22/2015

Totals =

Conch - 2012: 278 colonies, 2014: 38, Dry Rocks - 2012: 310, 2014: 151,
Molasses - 2012: 312, 2014: 126, Pickles - 2012: 304, 2014: 234

Maximum Depth: 9m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

The four sites shared ten genotypes. When analyzed by genotype in 2012, three general clusters appear: U1 was smaller than everything else, U3-U53-U54-U55-U61 was a middle grouping, and U17-U44-U51-U59 comprised the largest colonies (Table 2). By 2014, there were no differences in maximum skeletal diameter by genotype. Differences in average maximum skeletal diameter between 2012 and 2014 were statistically significant for all ten genotypes (Wilcoxon test $p < 0.0001$).

Table 2. Maximum skeletal diameter separated by genotype for 2012 projects, ranked by 2012 size. Letters indicate statistical significance within sampling.

| 2012 | | | | | 2014 | | |
|----------|----------------------------|---------|-----|------|----------------------------|---------|----|
| Genotype | Avg. Max. Skel. Diam. (cm) | SE (cm) | N | Sig. | Avg. Max. Skel. Diam. (cm) | SE (cm) | N |
| U44 | 18.56 | 0.62 | 100 | AB | 32.50 | 3.41 | 8 |
| U17 | 18.31 | 0.57 | 95 | B | 38.80 | 7.39 | 15 |
| U51 | 17.74 | 0.47 | 119 | AB | 37.33 | 2.78 | 15 |
| U59 | 17.63 | 0.59 | 122 | ABC | 32.50 | 2.88 | 13 |
| U3 | 16.60 | 0.45 | 123 | ABCD | 26.00 | 3.37 | 9 |
| U61 | 15.86 | 0.53 | 92 | ACD | 32.70 | 3.17 | 10 |
| U53 | 14.92 | 0.46 | 125 | CD | 32.18 | 1.99 | 11 |
| U55 | 14.79 | 0.52 | 120 | D | 34.56 | 3.22 | 9 |
| U54 | 14.18 | 0.38 | 74 | D | 33.18 | 4.78 | 11 |
| U1 | 12.32 | 0.33 | 120 | E | 26.82 | 2.80 | 11 |

When analyzed by habitat type, colonies transplanted to spur-and-groove habitats (Dry Rocks and Molasses Reef) were significantly larger than their counterparts in hard-bottom locations (Pickles and Conch Reefs) at the start of the project and in 2014 (Figure 49). At the beginning of the project, the average maximum skeletal diameter was 17.39 ± 0.25 cm for spur-and-groove colonies and 14.95 ± 0.19 cm for hard-bottom individuals (Wilcoxon $p < 0.0001$). By 2014, the average maximum skeletal diameter had increased to 31.81 ± 0.85 cm for spur-and-groove transplants and 28.56 ± 0.88 cm for hard-bottom colonies (Wilcoxon $p < 0.01$). Differences in average maximum skeletal diameter between 2012 and 2014 were statistically significant for both habitat types (Wilcoxon test $p < 0.0001$).

Maximum Skeletal Diameter vs. Time by Habitat Type 2012 Projects

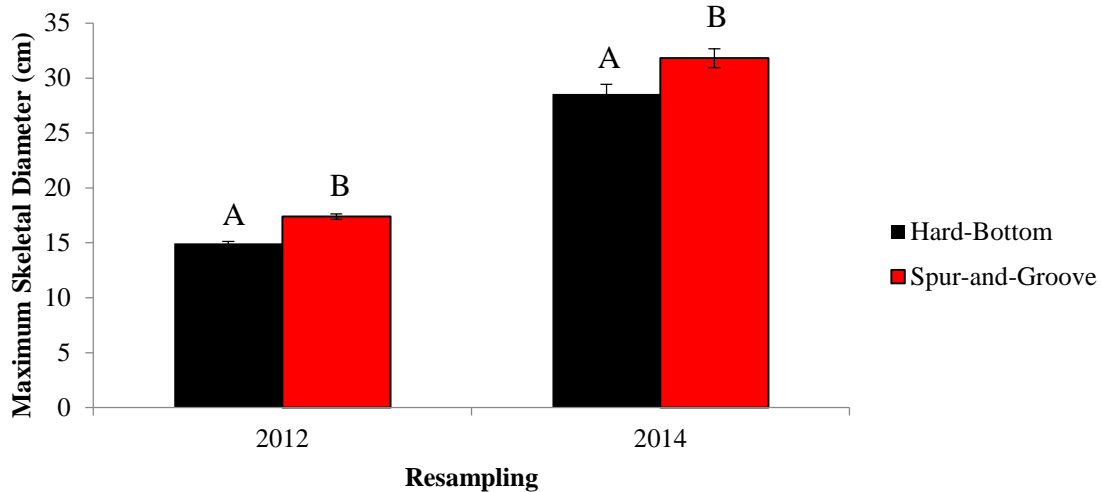


Figure 55. Maximum skeletal diameter by resampling separated by habitat type for 2012 projects.

Date Ranges = 2012: 4/24 - 6/6/2012, 2014: 10/29/2014 - 1/22/2015

Totals = Hard-bottom - 2012: 582 colonies, 2014: 272,

Spur-and-Groove - 2012: 622, 2014: 277

Maximum Depth: 9m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

Percent survival and percent live tissue coverage were significantly higher at Pickles Reef compared to the other three reefs (all Pairwise Wilcoxon $p < 0.0001$). Dry Rocks and Molasses Reef were significantly greater than Conch Reef (Pairwise Wilcoxon $p < 0.0001$). By 2014, colonies on Pickles Reef recorded an average survivorship of $72.00 \pm 2.49\%$ and $47.00 \pm 2.38\%$ live tissue coverage (Figures 50 and 51). Dry Rocks recorded $46.28 \pm 2.84\%$ percent survival with $27.38 \pm 2.20\%$ live tissue, while Molasses Reef had $40.91 \pm 2.81\%$ percent survival and $22.95 \pm 2.07\%$ live tissue. Conch Reef had the lowest observed survival and percent live tissue with $12.19 \pm 1.96\%$ survival and $5.71 \pm 1.27\%$ live tissue coverage. Differences in percent survival and percent live tissue between 2012 and 2014 were statistically significant for all four reefs (Wilcoxon test $p < 0.0001$).

Survival vs. Time by Reef 2012 Projects

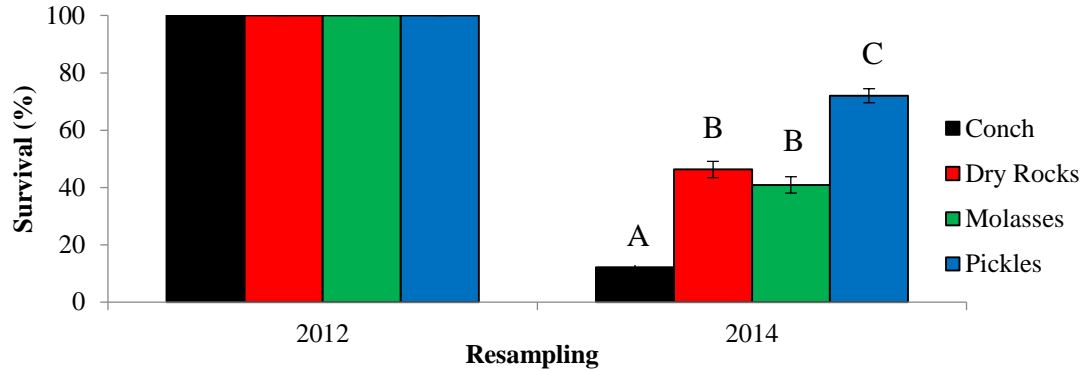


Figure 56. Colony survivorship by resampling separated by reef for 2012 projects.
 Date Ranges = 2012: 4/24 - 6/6/2012, 2014: 10/29/2014 - 1/22/2015
 Totals = Conch - 2012: 400 colonies, 2014: 290, Dry Rocks - 2012: 400, 2014: 318,
 Molasses - 2012: 400, 2014: 320, Pickles - 2012: 400, 2014: 325
 Maximum Depth: 9m
 Error bars indicate standard error. Letters indicate statistical significance within sampling.

Live Tissue vs. Time by Reef 2012 Projects

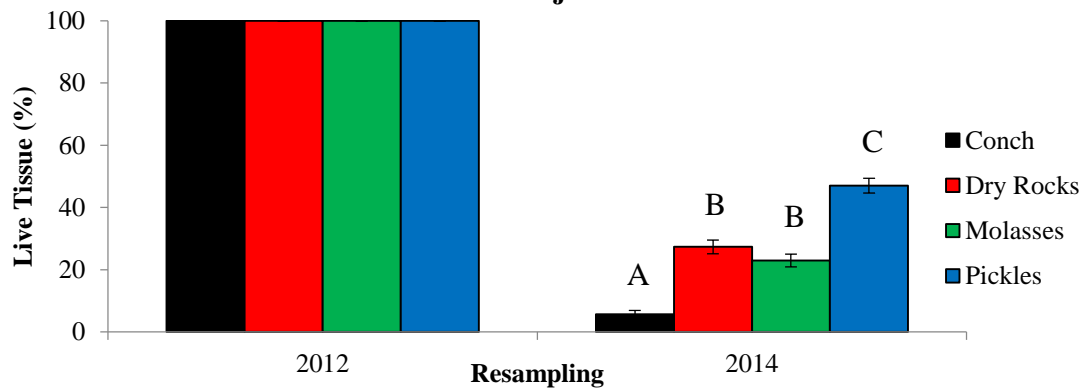


Figure 57. Percent live tissue by resampling separated by reef for 2012 projects.
 Date Ranges = 2012: 4/24 - 6/6/2012, 2014: 10/29/2014 - 1/22/2015
 Totals = Conch - 2012: 400 colonies, 2014: 290, Dry Rocks - 2012: 400, 2014: 318,
 Molasses - 2012: 400, 2014: 320, Pickles - 2012: 400, 2014: 325
 Maximum Depth: 9m
 Error bars indicate standard error. Letters indicate statistical significance within sampling.

When analyzed by genotype, no statistical differences in either percent survival or percent live tissue were observed combining results from all four reefs (Table 3). By 2014, many of the genotype tags had been lost. The small sample sizes resulted in high variance. Differences in percent survival and percent live tissue between 2012 and 2014 were statistically significant for all ten genotypes (Wilcoxon test $p < 0.0001$).

Table 3. Colony survivorship and percent live tissue separated by genotype for 2012 projects, ranked by percent survival.

| 2014 | | | | | | |
|----------|--------------|-----------|----|-----------------|-----------|----|
| Genotype | Survival (%) | | | Live Tissue (%) | | |
| | Average | Std Error | N | Average | Std Error | N |
| U51 | 88.24 | 0.81 | 17 | 44.82 | 10.21 | 17 |
| U54 | 84.62 | 1.04 | 13 | 71.92 | 9.94 | 13 |
| U3 | 81.82 | 1.25 | 11 | 33.64 | 10.70 | 11 |
| U59 | 81.25 | 1.01 | 16 | 32.75 | 9.09 | 16 |
| U53 | 78.57 | 1.11 | 14 | 41.93 | 10.04 | 14 |
| U1 | 73.33 | 1.18 | 15 | 35.07 | 10.79 | 15 |
| U17 | 71.43 | 1.01 | 21 | 41.48 | 9.43 | 21 |
| U61 | 71.43 | 1.25 | 14 | 39.14 | 10.89 | 14 |
| U55 | 64.29 | 1.33 | 14 | 28.29 | 9.49 | 14 |
| U44 | 57.14 | 1.37 | 14 | 21.07 | 8.58 | 14 |

When analyzed by habitat type, no statistical differences in survival or condition were found (Figures 52 and 53). By 2014, average survival at the hard-bottom sites was $44.37 \pm 2.02\%$ compared to $43.60 \pm 2.00\%$ at the spur-and-groove locations. Percent live tissue coverage was also similar, with hard-bottom colonies recording an average of $27.93 \pm 1.64\%$ compared to $25.17 \pm 1.51\%$ for those transplanted to spur-and-groove habitats. Differences in percent survival and percent live tissue between 2012 and 2014 were statistically significant for both habitat types (Wilcoxon test $p < 0.0001$).

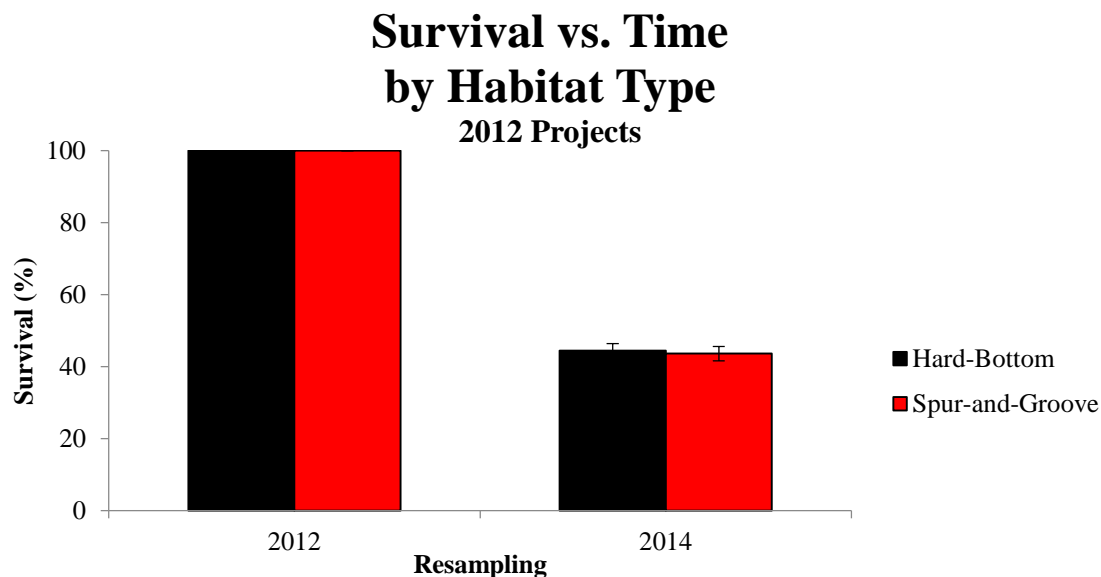


Figure 58. Colony survivorship by resampling separated by habitat type for 2012 projects.

Date Ranges = 2012: 4/24 - 6/6/2012, 2014: 10/29/2014 - 1/22/2015

Totals = Hard-bottom - 2012: 800 colonies, 2014: 615,

Spur-and-Groove - 2012: 800, 2014: 638

Maximum Depth: 9m

Error bars indicate standard error.

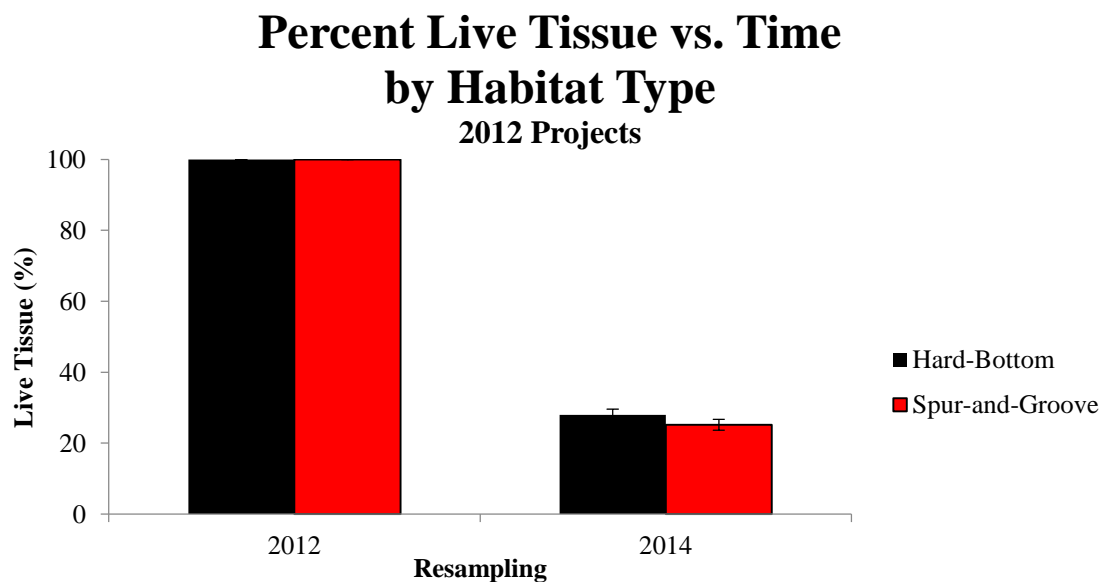


Figure 59. Percent live tissue by resampling separated by habitat type for 2012 projects.

Date Ranges = 2012: 4/24 - 6/6/2012, 2014: 10/29/2014 - 1/22/2015

Totals = Hard-bottom - 2012: 800 colonies, 2014: 615,

Spur-and-Groove - 2012: 800, 2014: 638

Maximum Depth: 9m

Error bars indicate standard error.

2013 PROJECTS

Three projects were conducted in 2013 – one on French Reef, one on Pickles Reef, and one on Conch Reef. For each project, colonies were deployed in clusters of approximately ten individuals from the same genotype. Small plastic tags indicating genotype were secured to a branch on one or two colonies within each cluster. A total of 29 genotypes were used, but none were repeated on all three reefs. The French project (project ID “FR2013”) was transplanted using 161 colonies on August 6, 2013, on a spur-and-groove structure at a depth of 10m under French Ball #4. 296 colonies at Pickles Reef (project ID “P2013”) were deployed between July 19 and September 10, 2013, at a depth of 7m on two spurs under Pickles Balls #1 and #2. Lastly, the 92 Conch Reef individuals (project ID “CNC2013”) were attached to a hard-bottom pavement habitat at a depth of 5m on December 17, 2013 under Conch Ball #2 in collaboration with The Nature Conservancy. No monitoring visits conducted by the Coral Restoration Foundation were included in the photographic record.

The average (\pm SE) initial maximum skeletal diameter for the 549 colonies recorded in the initial photos was 18.22 ± 0.31 cm. In 2014, the average maximum skeletal diameter increased to 29.04 ± 0.76 cm (Figure 60), an increase of 10.82 cm. The difference in average maximum skeletal diameter between 2013 and 2014 was statistically significant (Wilcoxon test $p \ll 0.0001$).

Maximum Skeletal Diameter vs. Time

2013 Projects

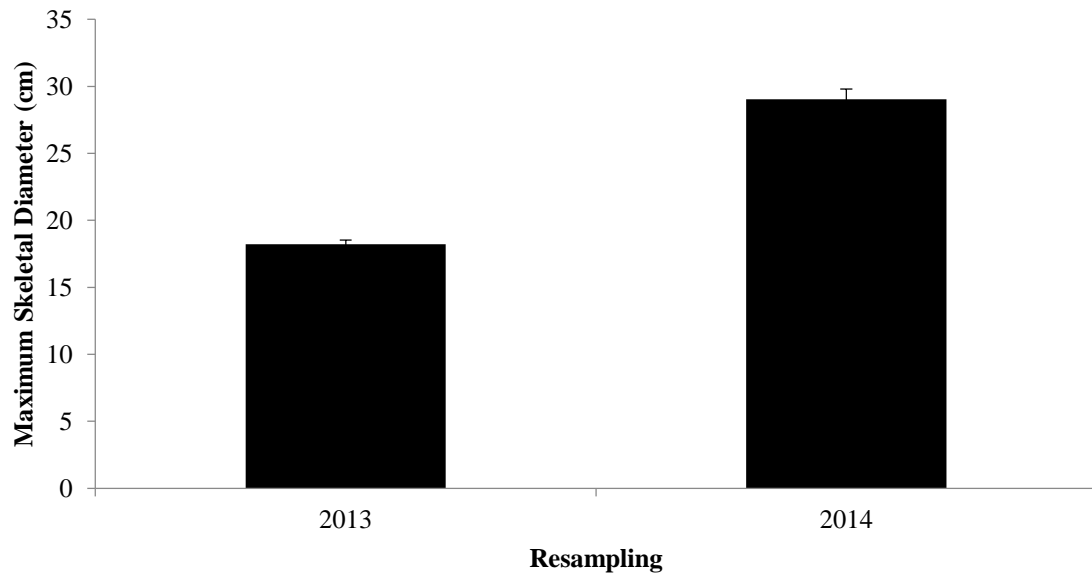


Figure 60. Maximum skeletal diameter by resampling for 2013 projects. Data averaged across all three reefs (Pickles Reef, Conch Reef, and French Reef). Date Ranges = 2013: 7/19 - 12/17/2012, 2014: 9/2/2014 - 1/21/2015
Totals = 2013: 549 colonies, 2014: 232
Maximum Depth: 10m
Error bars indicate standard error.

When analyzed by reef, colonies at French Reef were significantly larger at the initial transplanting compared to the other two sites (pairwise Wilcoxon $p < 0.0001$). In 2013, average maximum skeletal diameter was 15.71 ± 0.59 cm for Conch Reef, 20.62 ± 0.54 cm for French Reef, and 17.69 ± 0.44 cm for Pickles Reef (Figure 61). In 2014, the average maximum skeletal diameter increased to 29.12 ± 1.13 cm for Conch Reef, 25.01 ± 1.52 cm for French Reef, and 30.35 ± 1.08 cm for Pickles Reef. The difference between Conch Reef and French Reef was statistically significant (pairwise Wilcoxon $p < 0.01$). Differences in average maximum skeletal diameter between 2013 and 2014 were statistically significant for all three reefs (Wilcoxon test $p < 0.01$).

Maximum Skeletal Diameter vs. Time by Reef

2013 Projects

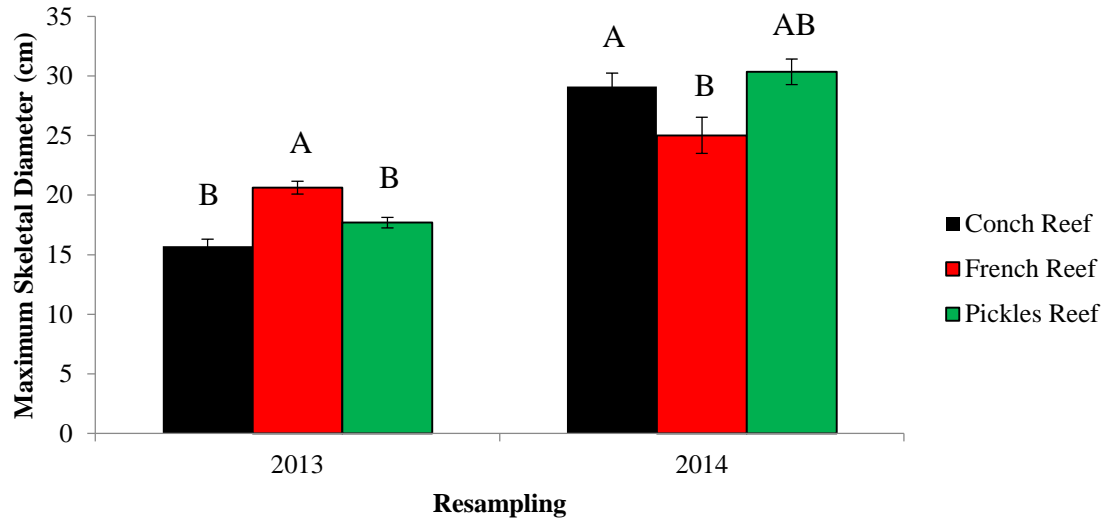


Figure 61. Maximum skeletal diameter by resampling separated by reef for 2013 projects. Date Ranges = 2013: 7/19 - 12/17/2012, 2014: 9/2/2014 - 1/21/2015

Totals =

Conch - 2013: 92 colonies, 2014: 68, French - 2013: 161, 2014: 72,

Pickles - 2013: 244, 2014: 217

Maximum Depth: 10m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

The three sites did not share any genotypes. The colonies displayed a large range in initial average skeletal diameter, from 28.65 ± 1.91 cm (U65) to 11.62 ± 0.53 (U51) (Table 4). In 2014, there were no differences in maximum skeletal diameter between genotypes. Missing genotype tags resulted in a reduced sample size. Of the 18 genotypes recorded in 2014, ten averaged above 30cm in diameter, four between 26cm and 30cm, and four below 26cm. Differences in average maximum skeletal diameter between 2013 and 2014 were statistically significant for 13 of the 18 genotypes (Wilcoxon test $p < 0.05$), including U12 which decreased in size. The differences in U5, U7, U52, U54, and U64 were not significant.

Table 4. Maximum skeletal diameter separated by genotype for 2013 projects, ranked by 2013 size. Letters indicate statistical significance within sampling.

| Genotype | 2013 | | | | 2014 | | |
|----------|----------------------------|---------|----|------------|----------------------------|---------|----|
| | Avg. Skel. Max. Diam. (cm) | SE (cm) | N | Signif. | Avg. Skel. Max. Diam. (cm) | SE (cm) | N |
| U65 | 28.65 | 1.91 | 7 | ADFGI | NA | | |
| U54 | 23.83 | 1.20 | 10 | ACDF | 26.50 | 7.24 | 4 |
| U70 | 22.02 | 1.29 | 44 | ACDE | 31.60 | 2.53 | 10 |
| U37 | 21.73 | 2.34 | 10 | ACDFH | NA | | |
| U59 | 21.72 | 1.31 | 36 | AE | 31.70 | 1.60 | 10 |
| U52 | 21.47 | 2.38 | 19 | AB | 30.64 | 5.67 | 14 |
| U4 | 20.82 | 1.17 | 27 | AE | 23.33 | 4.73 | 9 |
| U1 | 20.26 | 0.89 | 10 | ACDEF | 26.30 | 2.55 | 10 |
| U12 | 20.14 | 0.93 | 48 | ACE | 12.80 | 4.01 | 5 |
| U79 | 19.99 | 1.07 | 27 | AC | 31.68 | 1.86 | 19 |
| U7 | 19.31 | 1.24 | 25 | ACDEF G | 20.00 | 4.49 | 6 |
| U68 | 19.02 | 1.90 | 25 | AB | 34.00 | 10.69 | 3 |
| U20 | 18.55 | 0.89 | 26 | ACEF | 33.46 | 3.22 | 22 |
| U69 | 18.32 | 1.12 | 29 | ACDFG H | 26.00 | 1.92 | 4 |
| U63 | 18.15 | 1.58 | 9 | AB | NA | | |
| U15 | 17.06 | 2.02 | 23 | AB | NA | | |
| U24 | 16.96 | 2.03 | 16 | AB | NA | | |
| U32 | 16.37 | 2.29 | 9 | AB | NA | | |
| U56 | 15.22 | 1.34 | 8 | AB | 29.30 | 1.86 | 10 |
| U28 | 14.58 | 1.27 | 17 | AB | NA | | |
| U42 | 14.56 | 0.98 | 18 | BEH | NA | | |
| U22 | 13.79 | 1.29 | 10 | AB | NA | | |
| U53 | 13.67 | 1.29 | 15 | AB | 30.50 | 2.24 | 10 |
| U55 | 13.62 | 1.11 | 9 | AB | 32.00 | 4.40 | 6 |
| U21 | 13.40 | 0.67 | 17 | BG | NA | | |
| U25 | 12.15 | 0.78 | 26 | B | NA | | |
| U58 | 11.82 | 0.85 | 10 | BD | 30.11 | 2.56 | 9 |
| U64 | 11.77 | 0.83 | 9 | BCI | 17.00 | 5.86 | 7 |
| U51 | 11.62 | 0.53 | 10 | BFH | 32.38 | 1.78 | 8 |

When analyzed by habitat type, colonies transplanted to spur-and-groove habitats (French Reef and Pickles Reef) were significantly larger than their counterparts in hard-bottom locations (Conch Reef) at the start of the project in 2013 (Figure 62). At the

beginning of the project, the average maximum skeletal diameter was 18.73 ± 0.35 cm for spur-and-groove colonies and 15.71 ± 0.59 cm for hard-bottom individuals (Wilcoxon $p < 0.0001$). By 2014, the average maximum skeletal diameter had increased to 29.02 ± 0.90 cm for spur-and-groove transplants and 29.56 ± 1.12 cm for hard-bottom colonies (Wilcoxon $p = 0.09$). Differences in average maximum skeletal diameter between 2013 and 2014 were statistically significant for both habitat types (Wilcoxon test $p < 0.0001$).

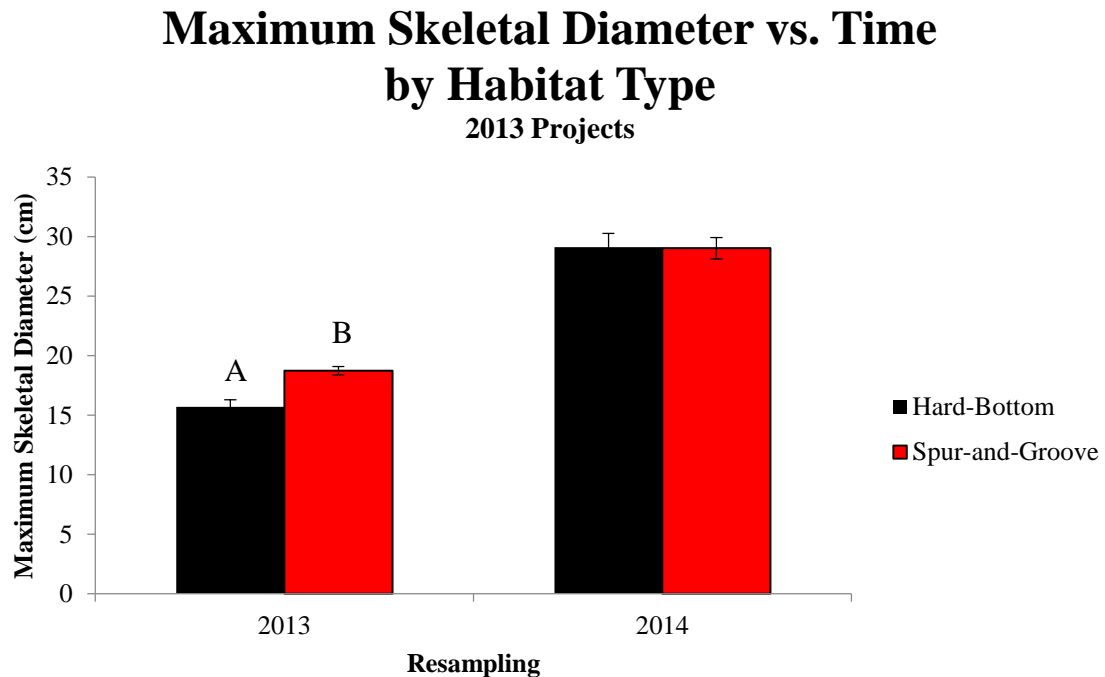


Figure 62. Maximum skeletal diameter by resampling separated by habitat type for 2013 projects.

Date Ranges = 2013: 7/19 - 12/17/2012, 2014: 9/2/2014 - 1/21/2015

Totals = Hard-bottom - 2013: 92 colonies, 2014: 68,

Spur-and-Groove - 2013: 457, 2014: 289

Maximum Depth: 10m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

Percent survival was significantly greater at Conch Reef compared to French Reef in 2014 (Pairwise Wilcoxon $p < 0.05$). No significant difference was observed between the reefs in percent live tissue in 2014. Colonies on Pickles Reef recorded an average survivorship of $69.78 \pm 2.61\%$ and $60.54 \pm 2.51\%$ live tissue coverage (Figures 63 and 64). Conch Reef recorded $81.93 \pm 4.25\%$ percent survival with $57.95 \pm 4.44\%$ live tissue, while French Reef had $64.87 \pm 4.55\%$ percent survival and $52.52 \pm 4.22\%$ live tissue.

Differences in percent survival and percent live tissue between 2013 and 2014 were statistically significant for all three reefs (all Wilcoxon test $p < 0.0001$).

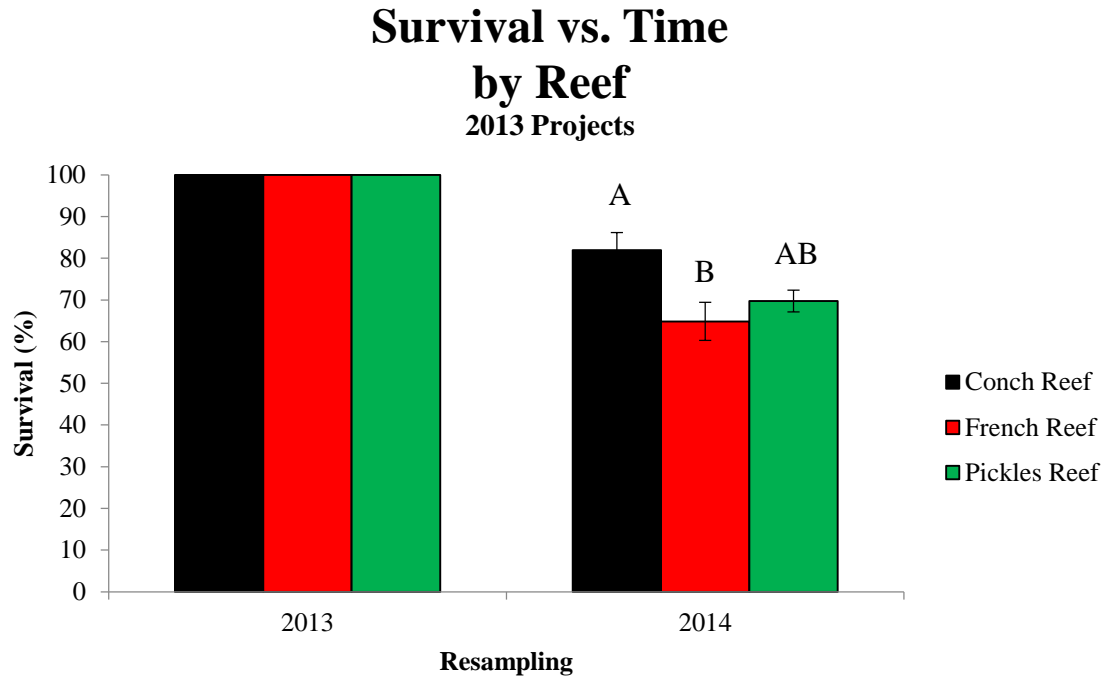


Figure 63. Colony survivorship by resampling separated by reef for 2013 projects.

Date Ranges = 2013: 7/19 - 12/17/2012, 2014: 9/2/2014 - 1/21/2015

Totals = Conch - 2013: 92 colonies, 2014: 83, French - 2013: 161, 2014: 111,

Pickles - 2013: 296, 2014: 311

Maximum Depth: 10m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

Percent Live Tissue vs. Time by Reef

2013 Projects

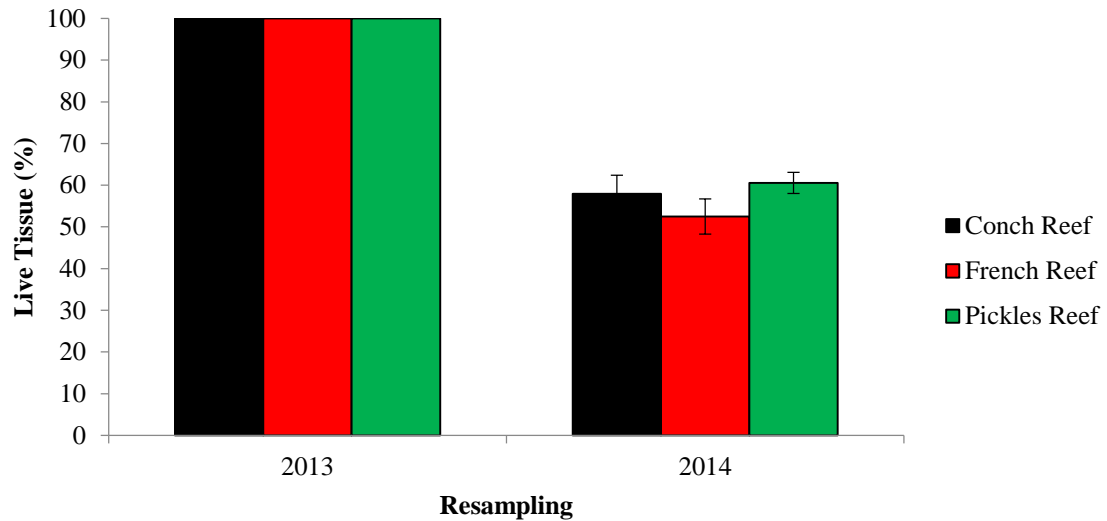


Figure 64. Percent live tissue by resampling separated by reef for 2013 projects.

Date Ranges = 2013: 7/19 - 12/17/2012, 2014: 9/2/2014 - 1/21/2015

Totals = Conch - 2013: 92 colonies, 2014: 83, French - 2013: 161, 2014: 111,

Pickles - 2013: 296, 2014: 311

Maximum Depth: 10m

Error bars indicate standard error. Letters indicate statistical significance within sampling.

When analyzed by genotype, several statistical differences in both percent survival and percent live tissue were observed in 2014 (Table 5). U20 colonies had significantly greater percent survival than those from U55, U68, and U70 (pairwise Wilcoxon $p < 0.05$ with Bonferroni adjustment). These colonies also had greater percent live tissue than those from U51, U53, U54, U55, and U79 (pairwise Wilcoxon $p < 0.05$ with Bonferroni adjustment). Colonies from U53 had less live tissue than U52 and U56 as well. Many of the genotype tags had been lost by 2014, resulting in 11 of the 29 genotypes missing from the analysis. Differences in percent survival between 2013 and 2014 were statistically significant for genotypes U4, U7, U12, U54, U55, U68, U69, U70, and U79 (Wilcoxon test $p < 0.0001$). Differences in percent live tissue between 2013 and 2014 were statistically significant for genotypes for all of the genotypes recorded in 2014 except U1 and U64 (Wilcoxon test $p < 0.05$).

Table 5. Colony survivorship and percent live tissue separated by genotype for 2013 projects, ranked by 2014 percent survival. Letters indicate statistical significance within sampling.

| 2014 | | | | | | | | |
|------|--------------|------------|----|---------|-----------------|------------|----|---------|
| | Survival (%) | | | | Live Tissue (%) | | | |
| | Average | Std. Error | N | Signif. | Average | Std. Error | N | Signif. |
| U20 | 100 | 0 | 22 | A | 94.5 | 1.966 | 22 | A |
| U51 | 100 | 0 | 8 | AB | 58.125 | 10.562 | 8 | BC |
| U52 | 100 | 0 | 14 | AB | 83.929 | 6.337 | 14 | AB |
| U56 | 100 | 0 | 10 | AB | 95.2 | 1.837 | 10 | AB |
| U58 | 90 | 10 | 10 | AB | 71 | 11.757 | 10 | ABC |
| U1 | 83.333 | 11.237 | 12 | AB | 79.583 | 11.12 | 12 | ABC |
| U59 | 83.333 | 11.237 | 12 | AB | 65.667 | 11.777 | 12 | ABC |
| U64 | 77.778 | 14.699 | 9 | AB | 71.667 | 14.577 | 9 | ABC |
| U53 | 76.923 | 12.163 | 13 | AB | 26.538 | 8.405 | 13 | C |
| U12 | 71.429 | 18.443 | 7 | AB | 61.429 | 17.786 | 7 | ABC |
| U7 | 66.667 | 16.667 | 9 | AB | 61.111 | 15.406 | 9 | ABC |
| U79 | 63.333 | 8.949 | 30 | AB | 47.7 | 8.047 | 30 | BC |
| U4 | 60 | 13.093 | 15 | AB | 48 | 11.596 | 15 | ABC |
| U54 | 50 | 18.898 | 8 | AB | 30 | 14.299 | 8 | BC |
| U69 | 50 | 18.898 | 8 | AB | 48.375 | 18.292 | 8 | ABC |
| U70 | 43.478 | 10.569 | 23 | B | 39.348 | 9.797 | 23 | ABC |
| U68 | 42.857 | 20.203 | 7 | B | 42.857 | 20.203 | 7 | ABC |
| U55 | 40 | 13.093 | 15 | B | 27.667 | 11.158 | 15 | BC |

When analyzed by habitat type, a statistically significant difference in percent survival was found, but not in percent live tissue (Figures 65 and 66). By 2014, average survival at the hard-bottom sites was $81.93 \pm 4.25\%$ compared to $68.48 \pm 2.26\%$ at the spur-and-groove locations. Percent live tissue coverage in hard-bottom colonies was $57.95 \pm 4.44\%$ compared to $58.43 \pm 2.16\%$ for those transplanted to spur-and-groove habitats. Differences in percent survival and percent live tissue between 2013 and 2014 were statistically significant for both habitat types (all Wilcoxon test $p < 0.0001$).

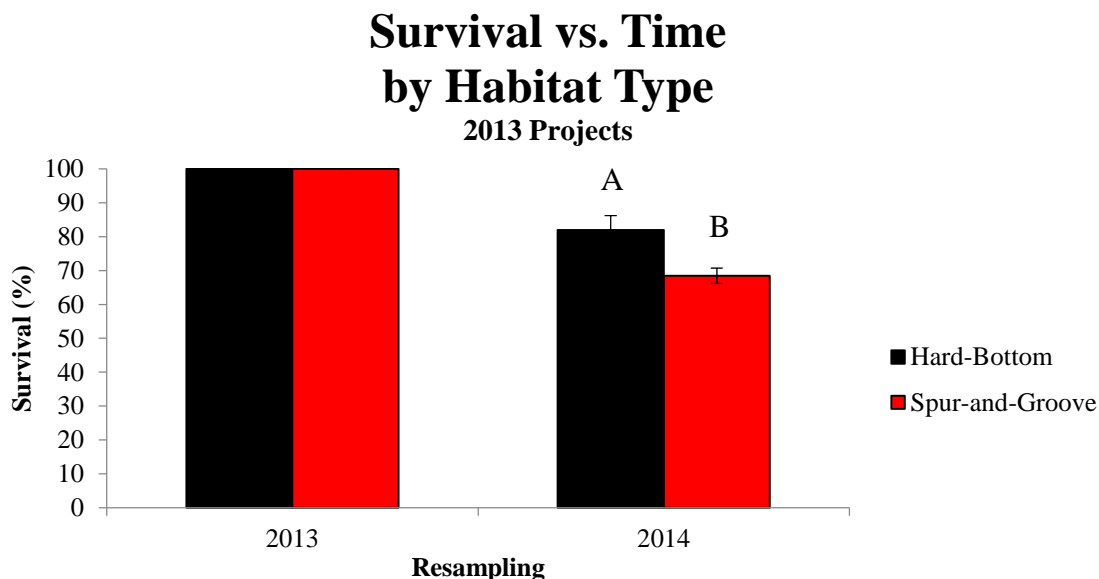


Figure 65. Colony survivorship by resampling separated by habitat type for 2013 projects.

Date Ranges = 2013: 7/19 - 12/17/2012, 2014: 9/2/2014 - 1/21/2015

Totals = Hard-bottom - 2013: 92 colonies, 2014: 83,

Spur-and-Groove - 2013: 457, 2014: 422

Maximum Depth: 10m

Error bars indicate standard error.

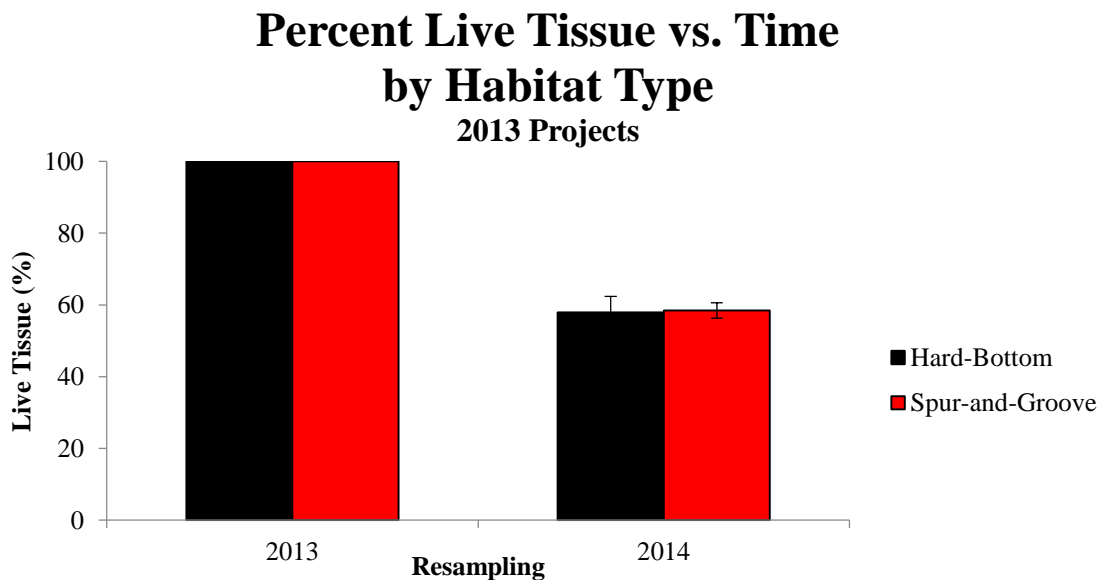


Figure 66. Percent live tissue by resampling separated by habitat type for 2013 projects.

Date Ranges = 2013: 7/19 - 12/17/2012, 2014: 9/2/2014 - 1/21/2015

Totals = Hard-bottom - 2013: 92 colonies, 2014: 83,

Spur-and-Groove - 2013: 457, 2014: 422

Maximum Depth: 10m

Error bars indicate standard error.

LITTLE CONCH REEF EXPERIMENT

OVERALL

Results are first presented combining colonies from both shallow and deep sites, without distinguishing between clusters or thickets, and before stacking occurred. After that, results are broken down by site, transplant structure (clusters and thickets), and genotype. The average (\pm SE) maximum skeletal diameter of all non-stacked colonies deployed at the Little Conch site after approximately one month on the reef was 23.78 ± 0.44 cm. After the 1.2 year-long study, the average increased to 36.04 ± 0.99 cm, an increase of 12.26 cm (Table 6, Wilcoxon $p \ll 0.0001$). Growth appeared linear with an average increase of 11.83 cm yr^{-1} (Figure 61). By 2015, percent survival had fallen to $28.67 \pm 1.90\%$, and of those still living, average live tissue coverage was only $10.12 \pm 0.98\%$ (Figures 62 & 63, both Wilcoxon $p \ll 0.0001$).

Table 6: Little Conch maximum skeletal diameter averaged by resampling effort. Only non-stacked structures from both sites are included.

| Sampling | # Days Deployed | Average Maximum Skeletal Diameter (cm) | Standard Error (cm) | Sample Size |
|-------------|-----------------|--|---------------------|-------------|
| Winter 2013 | 47 | 23.78 | 0.44 | 817 |
| Summer 2014 | 207 | 28.97 | 0.48 | 728 |
| Autumn 2014 | 329 | 33.74 | 0.77 | 340 |
| Winter 2015 | 435 | 36.04 | 0.99 | 168 |

Maximum Skeletal Diameter vs. Time

Overall

Little Conch Reef

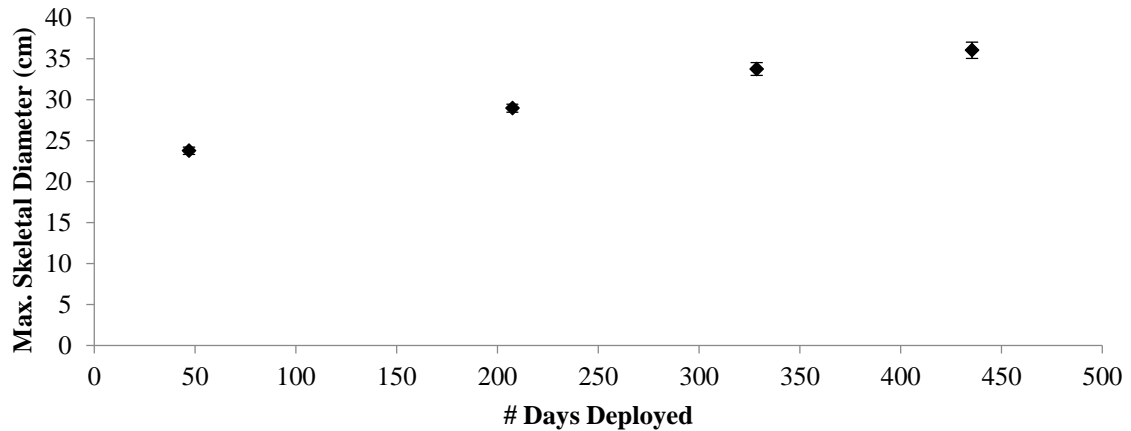


Figure 67: Little Conch maximum skeletal diameter vs. number of days deployed. Only non-stacked structures from both sites are included.
 Winter 2013: 817 colonies, Summer 2014: 728, Autumn 2014: 340, Winter 2015: 168
 Deep site: 12 m, Shallow site: 5 m
 Error bars indicate standard error.
 Average Growth Rate = 11.83 cm yr⁻¹

Survival vs. Time

Overall

Little Conch Reef

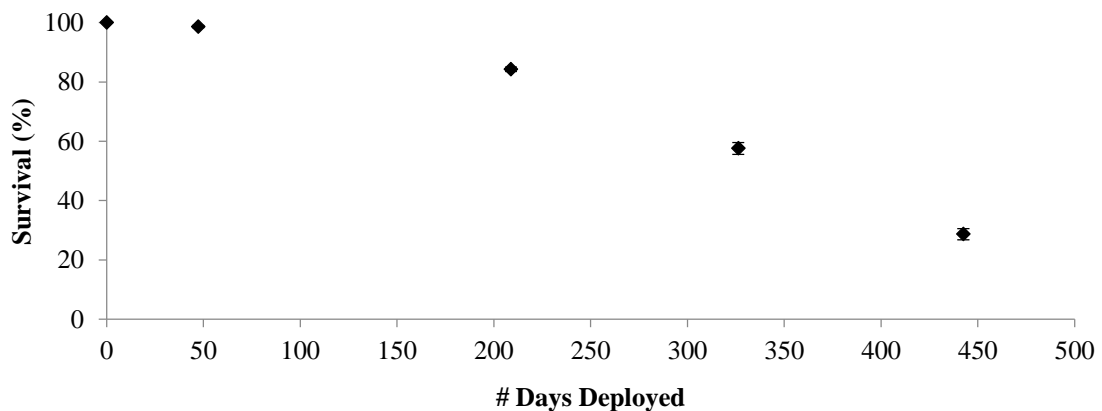


Figure 68: Little Conch percent survival vs. number of days deployed. Only non-stacked structures from both sites are included.
 Percent survival = ((# alive/# total)*100)
 Initial transplant: 1288 colonies, Winter 2013: 828, Summer 2014: 864, Autumn 2014: 641, Winter 2015: 586
 Error bars indicate standard error.

Live Tissue vs. Time Overall Little Conch Reef

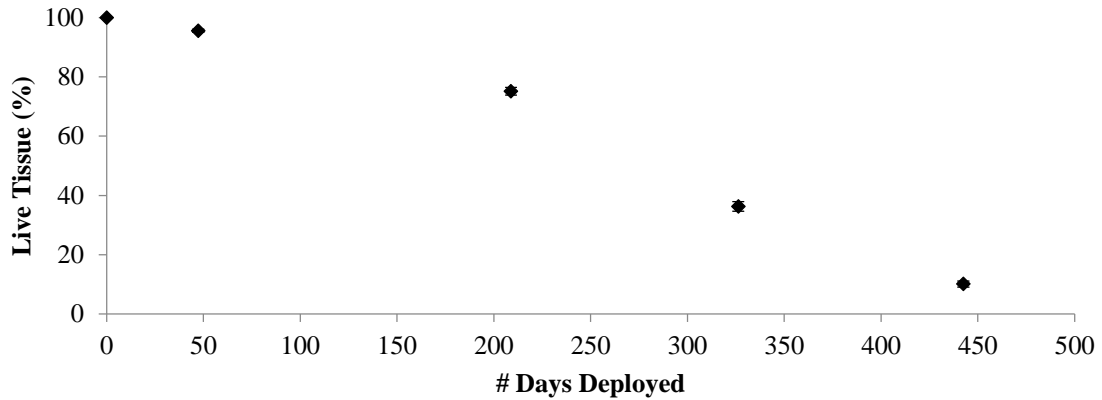


Figure 69: Little Conch percent live tissue vs. number of days deployed. Only non-stacked structures from both sites are included. Initial transplant: 1288 colonies, Winter 2013: 828, Summer 2014: 864, Autumn 2014: 612, Winter 2015: 586
Error bars indicate standard error.

SHALLOW VS. DEEP

At the Shallow (5m) site, the average (\pm SE) maximum skeletal diameter of the colonies deployed after approximately one month on the reef was 22.39 ± 0.53 cm. After over 400 days on the reef, the average had increased to 35.41 ± 1.10 cm, an increase of 13.02 cm (Table 7). Growth appeared linear with an average increase of 12.15 cm yr^{-1} (Figure 64). By 2015, percent survival had fallen to $65.05 \pm 3.33\%$, and of those still living, average live tissue coverage was only $26.30 \pm 2.31\%$ (Figures 65 and 66).

Average maximum skeletal diameter at the Deep (12m) site increased from 25.80 ± 0.76 cm to 38.50 ± 2.19 cm (Table 7). Over the course of the study, percent survival fell to $8.95 \pm 1.47\%$ (Figure 65). Little tissue remained on the living colonies by the Winter 2015 resampling ($1.35 \pm 0.39\%$, Figure 66).

Table 7: Maximum skeletal diameter separated by resampling effort and depth. Non-stacked structures only.

| Site | Sampling | # Days Deployed | Average Skeletal Maximum Diameter (cm) | Standard Error (cm) | Sample Size |
|--------------|-------------|-----------------|--|---------------------|-------------|
| Shallow (5m) | Winter 2013 | 31 | 22.39 | 0.53 | 485 |
| | Summer 2014 | 201 | 27.60 | 0.53 | 520 |
| | Autumn 2014 | 334 | 33.02 | 0.81 | 239 |
| | Winter 2015 | 433 | 35.41 | 1.10 | 134 |
| Deep (12m) | Winter 2013 | 71 | 25.80 | 0.76 | 332 |
| | Summer 2014 | 208 | 32.38 | 1.03 | 208 |
| | Autumn 2014 | 316 | 35.46 | 1.72 | 101 |
| | Winter 2015 | 445 | 38.50 | 2.19 | 34 |

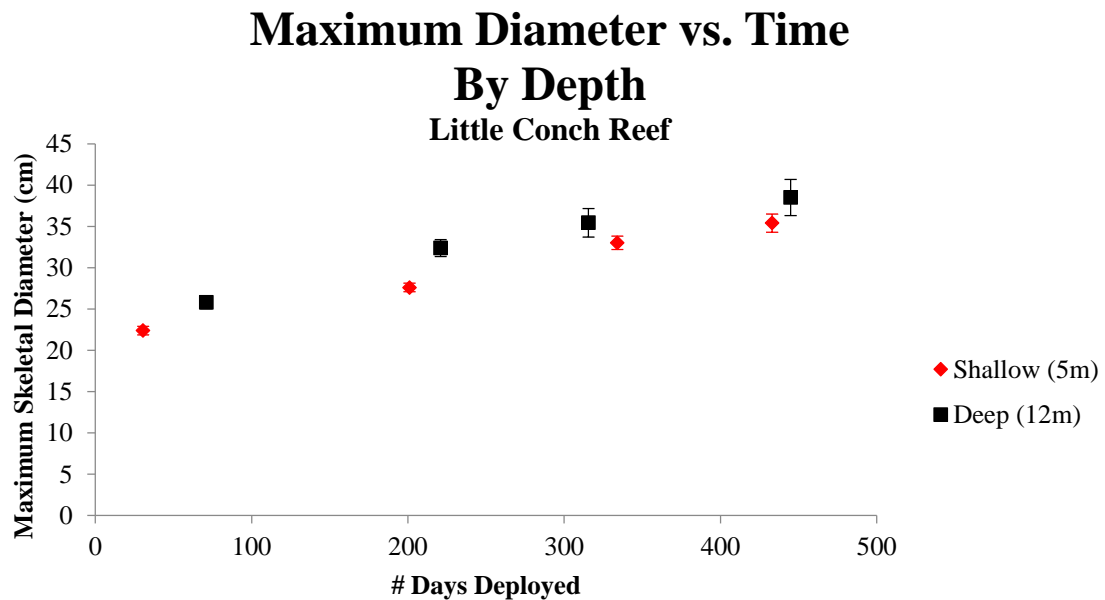


Figure 70: Maximum skeletal diameter vs. number of days deployed separated by depth. Non-stacked structures only.

Shallow totals: Winter 2013: 485 colonies, Summer 2014: 520, Autumn 2014: 239, Winter 2015: 134

Deep totals: Winter 2013: 332 colonies, Summer 2014: 208, Autumn 2014: 101, Winter 2015: 34

Deep site: 12m, Shallow site: 5m

Error bars indicate standard error.

Survival vs. Time By Depth Little Conch Reef

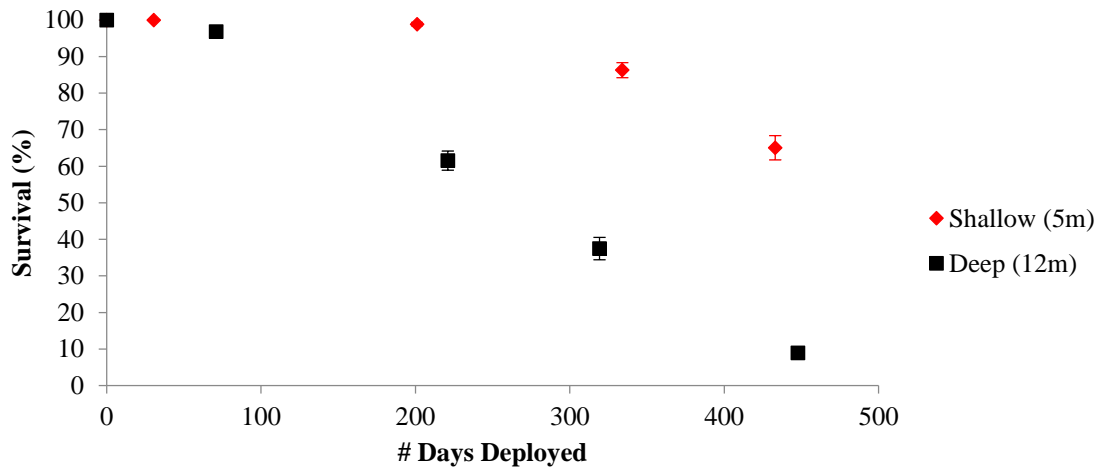


Figure 71: Percent survival vs. number of days deployed separated by depth.

Non-stacked structures only.

Percent survival = $((\# \text{ alive} / \# \text{ total}) * 100)$

Shallow (5m) site: Initial transplant: 644 colonies, Winter 2013: 485, Summer 2014: 526, Autumn 2014: 277, Winter 2015: 206

Deep (12m) site: Initial transplant: 644 colonies, Winter 2013: 343, Summer 2014: 338, Autumn 2014: 248, Winter 2015: 380

Error bars indicate standard error.

Live Tissue vs. Time By Depth Little Conch Reef

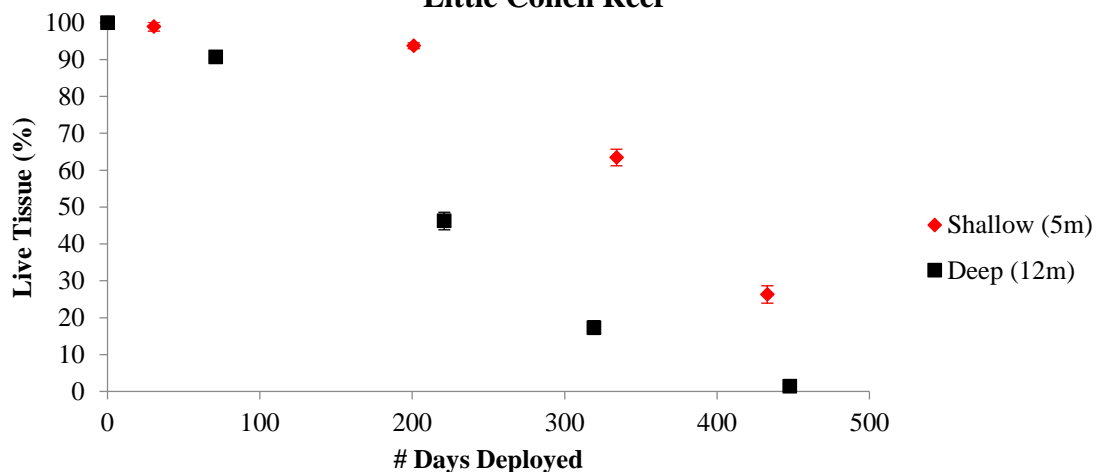


Figure 72: Percent live tissue vs. number of days deployed separated by depth.

Non-stacked structures only.

Shallow (5m) site: Initial transplant: 644 colonies, Winter 2013: 485, Summer 2014: 526, Autumn 2014: 277, Winter 2015: 206

Deep (12m) site: Initial transplant: 644 colonies, Winter 2013: 343, Summer 2014: 338, Autumn 2014: 248, Winter 2015: 380

Error bars indicate standard error.

Wilcoxon tests revealed no difference in average skeletal diameter by depth at the study's conclusion ($p = 0.06107$). Differences in percent survival and live tissue coverage were significant (both $p < 0.0001$). Colonies at the shallow site demonstrated both greater percent survival and percent live tissue coverage than colonies at the deep site.

CLUSTER VS. THICKET

At the shallow (5m) site, the average (\pm SE) maximum skeletal diameter of colonies deployed in clusters increased from 21.63 ± 1.02 cm in Winter 2013 to 34.65 ± 1.76 cm in Winter 2015 (Table 8). By 2015, percent survival in these structures had declined to $68.75 \pm 5.21\%$ with an average percent live tissue of $40.89 \pm 4.40\%$ (Figures 67 & 68). Colonies in thickets increased from 22.84 ± 0.59 cm to 35.94 ± 1.42 cm. Percent survival decreased to $62.70 \pm 4.33\%$ while percent live tissue coverage decreased to $17.04 \pm 2.19\%$.

Table 8: Maximum skeletal diameter separated by resampling effort and structure for shallow (5m) site. Non-stacked structures only.

| Structure | Sampling | # Days Deployed | Average Skeletal Maximum Diameter (cm) | Standard Error (cm) | Sample Size |
|-----------|-------------|-----------------|--|---------------------|-------------|
| Cluster | Winter 2013 | 20 | 21.63 | 1.02 | 180 |
| | Summer 2014 | 201 | 26.96 | 0.82 | 189 |
| | Autumn 2014 | 334 | 32.55 | 1.62 | 66 |
| | Winter 2015 | 433 | 34.65 | 1.76 | 55 |
| Thicket | Winter 2013 | 37 | 22.84 | 0.59 | 305 |
| | Summer 2014 | 201 | 27.97 | 0.68 | 331 |
| | Autumn 2014 | 334 | 33.20 | 0.93 | 173 |
| | Winter 2015 | 433 | 35.94 | 1.42 | 79 |

Survival vs. Time By Depth and Structure

Little Conch Reef

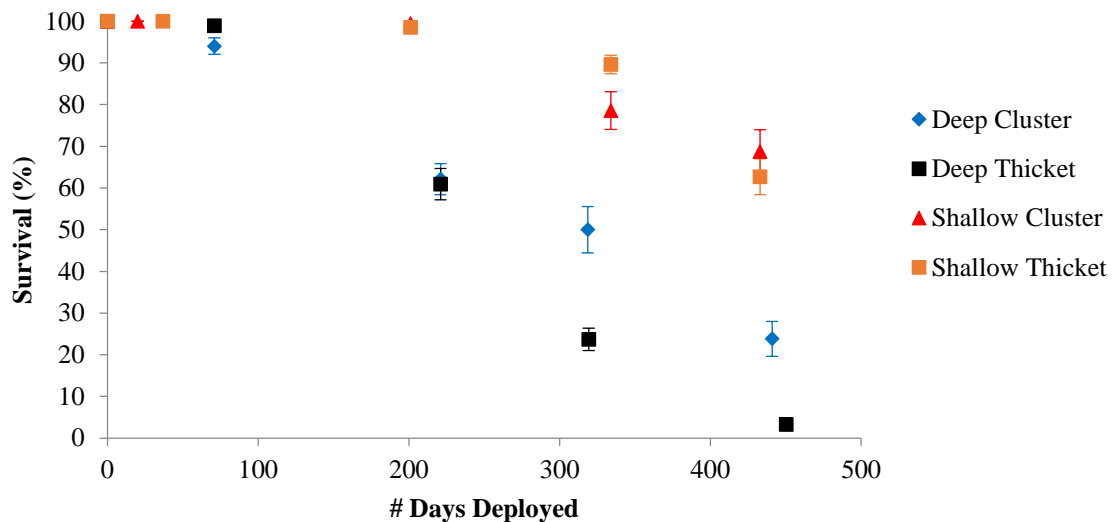


Figure 73: Percent survival vs. number of days deployed separated by depth and structure.

Non-stacked structures only.

Percent survival = $((\# \text{ alive} / \# \text{ total}) * 100)$

Shallow (5m) clusters: Initial transplant: 224 colonies, Winter 2013: 180, Summer 2014: 190, Autumn 2014: 84, Winter 2015: 80

Shallow (5m) thickets: Initial transplant: 420 colonies, Winter 2013: 305, Summer 2014: 336, Autumn 2014: 193, Winter 2015: 126

Deep (12m) clusters: Initial transplant: 224 colonies, Winter 2013: 151, Summer 2014: 169, Autumn 2014: 82, Winter 2015: 105

Deep (12m) thickets: Initial transplant: 420 colonies, Winter 2013: 192, Summer 2014: 169, Autumn 2014: 253, Winter 2015: 275

Error bars indicate standard error.

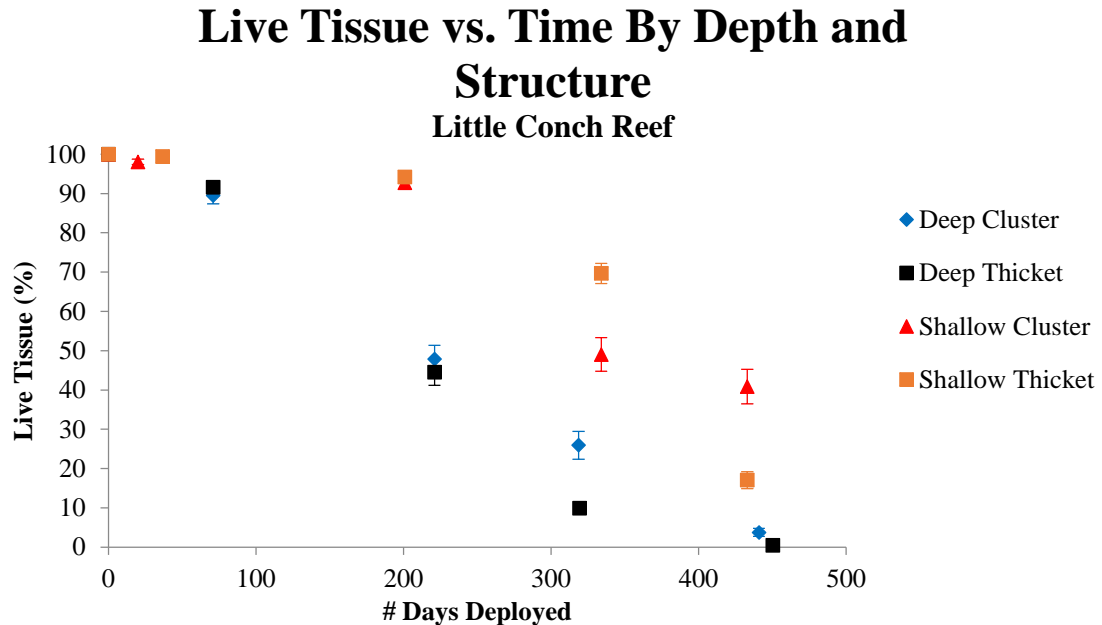


Figure 74: Percent live tissue vs. number of days deployed separated by depth and structure.

Non-stacked structures only.

Shallow (5m) clusters: Initial transplant: 224 colonies, Winter 2013: 180, Summer 2014: 190, Autumn 2014: 84, Winter 2015: 80

Shallow (5m) thickets: Initial transplant: 420 colonies, Winter 2013: 305, Summer 2014: 336, Autumn 2014: 193, Winter 2015: 126

Deep (12m) clusters: Initial transplant: 224 colonies, Winter 2013: 151, Summer 2014: 169, Autumn 2014: 82, Winter 2015: 105

Deep (12m) thickets: Initial transplant: 420 colonies, Winter 2013: 192, Summer 2014: 169, Autumn 2014: 253, Winter 2015: 275

Error bars indicate standard error.

Differences in maximum skeletal diameter and percent survival between clusters and thickets were not statistically significant at the shallow (5m) site. For percent live tissue coverage, colonies within the thickets had less live tissue than those in clusters ($p < 0.001$).

At the Deep (12m) site, average (\pm SE) maximum skeletal diameter of colonies contained in clusters increased by almost 13 cm in just over one year, from 25.37 ± 0.97 cm to 38.20 ± 1.47 cm (Table 9). In the same time period, percent survival fell to $23.81 \pm 4.18\%$ while percent live tissue coverage declined to $3.73 \pm 1.01\%$ (Figures 67 & 68). Colonies in the thickets increased in size from 26.12 ± 1.12 cm in 2013 to 39.33 ± 7.55 cm in 2015. By the end of the study, only $3.27 \pm 1.07\%$ of the colonies in these structures had living tissue with an average live tissue coverage of just $0.44 \pm 0.37\%$.

Table 9: Maximum skeletal diameter separated by resampling effort and structure for deep (12m) site. Non-stacked structures only.

| Structure | Sampling | # Days Deployed | Average Skeletal Maximum Diameter (cm) | Standard Error (cm) | Sample Size |
|-----------|-------------|-----------------|--|---------------------|-------------|
| Cluster | Winter 2013 | 71 | 25.37 | 0.97 | 142 |
| | Summer 2014 | 221 | 32.12 | 1.19 | 105 |
| | Autumn 2014 | 318 | 40.12 | 3.33 | 41 |
| | Winter 2015 | 440 | 38.20 | 1.47 | 25 |
| Thicket | Winter 2013 | 71 | 26.12 | 1.12 | 190 |
| | Summer 2014 | 221 | 32.65 | 1.70 | 103 |
| | Autumn 2014 | 314 | 32.27 | 1.70 | 60 |
| | Winter 2015 | 458 | 39.33 | 7.55 | 9 |

No significant differences in maximum skeletal diameter were found related to transplant structure at the deep site. However, clusters had greater percent survival and live tissue coverage than thickets ($p \ll 0.0001$ for both).

MULTIGENETIC CLUSTER VS. MONOGENETIC CLUSTER

The average (\pm SE) maximum skeletal diameter of colonies in multigenetic (“Mix”) clusters at the shallow (5m) site increased from 22.94 ± 0.89 cm in Winter 2013 to 32.55 ± 1.51 cm in Winter 2015 (Table 10). By 2015, percent survival in these structures had declined to $86.11 \pm 5.85\%$ with an average percent live tissue of $57.39 \pm 6.78\%$ (Figures 69 & 70). Colonies in monogenetic (“Single”) clusters increased from 20.49 ± 1.74 cm to 37.38 ± 3.50 cm. Percent survival decreased to $54.55 \pm 7.59\%$ while percent live tissue coverage fell to $27.39 \pm 4.97\%$.

Table 10: Maximum skeletal diameter separated by resampling effort and genetic composition for shallow (5m) site clusters. Non-stacked structures only.

| Genetic Comp. | Sampling | # Days Deployed | Average Skeletal Maximum Diameter (cm) | Standard Error (cm) | Sample Size |
|------------------------|-------------|-----------------|--|---------------------|-------------|
| Multi-genetic Clusters | Winter 2013 | 20 | 22.94 | 0.89 | 84 |
| | Summer 2014 | 201 | 28.39 | 0.98 | 67 |
| | Autumn 2014 | 334 | 33.28 | 2.10 | 32 |
| | Winter 2015 | 433 | 32.55 | 1.51 | 31 |
| Mono-genetic Clusters | Winter 2013 | 20 | 20.49 | 1.74 | 96 |
| | Summer 2014 | 201 | 26.18 | 1.15 | 122 |
| | Autumn 2014 | 334 | 31.85 | 2.46 | 34 |
| | Winter 2015 | 433 | 37.38 | 3.50 | 24 |

Survival vs. Time By Depth and Composition

Little Conch Reef

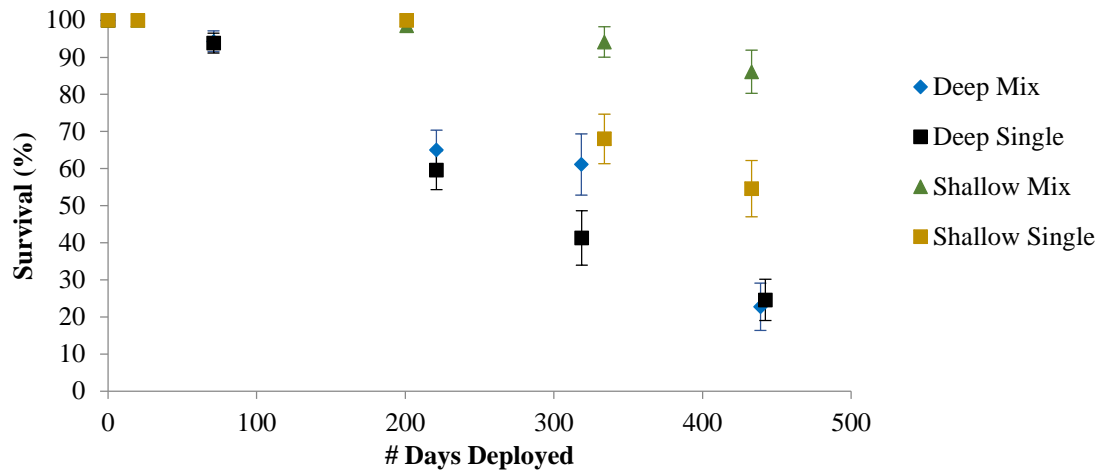


Figure 75: Percent survival vs. number of days deployed separated by depth and genetic composition

Non-stacked clusters only.

Percent survival = $((\# \text{ alive} / \# \text{ total}) * 100)$

Shallow (5m) mix: Initial transplant: 84 colonies, Winter 2013: 84, Summer 2014: 68, Autumn 2014: 34, Winter 2015: 36

Shallow (5m) single: Initial transplant: 140 colonies, Winter 2013: 96, Summer 2014: 122, Autumn 2014: 50, Winter 2015: 44

Deep (12m) mix: Initial transplant: 84 colonies, Winter 2013: 70, Summer 2014: 80, Autumn 2014: 36, Winter 2015: 44

Deep (12m) single: Initial transplant: 140 colonies, Winter 2013: 81, Summer 2014: 89, Autumn 2014: 46, Winter 2015: 61

Error bars indicate standard error.

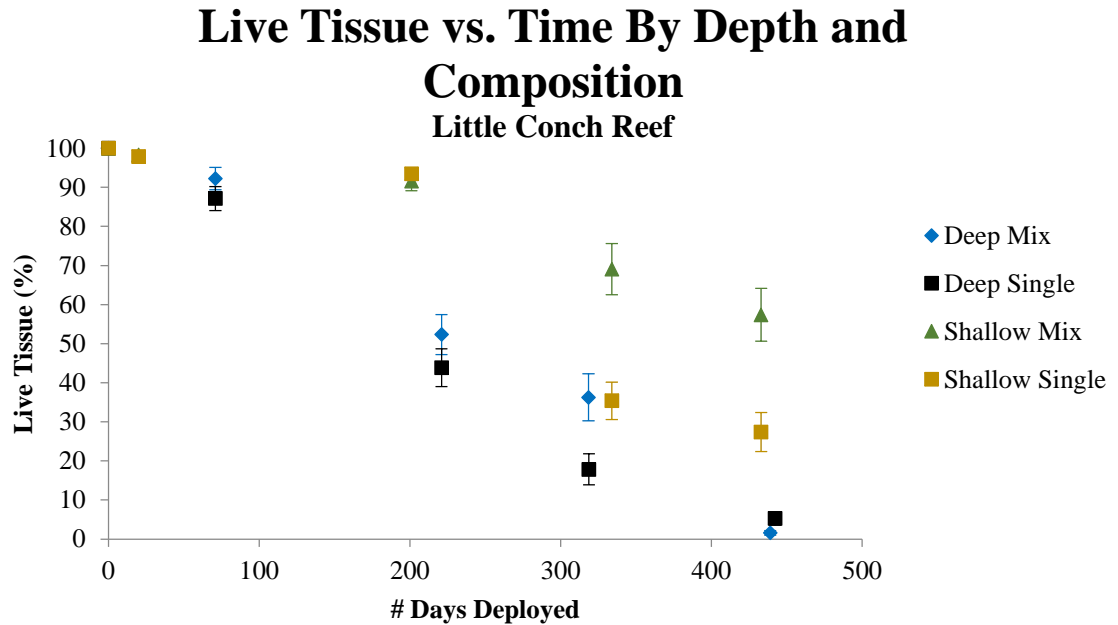


Figure 76: Percent live tissue vs. number of days deployed separated by depth and genetic composition.

Non-stacked clusters only.

Shallow (5m) mix: Initial transplant: 84 colonies, Winter 2013: 84, Summer 2014: 68, Autumn 2014: 34, Winter 2015: 36

Shallow (5m) single: Initial transplant: 140 colonies, Winter 2013: 96, Summer 2014: 122, Autumn 2014: 50, Winter 2015: 44

Deep (12m) mix: Initial transplant: 84 colonies, Winter 2013: 70, Summer 2014: 80, Autumn 2014: 36, Winter 2015: 44

Deep (12m) single: Initial transplant: 140 colonies, Winter 2013: 81, Summer 2014: 89, Autumn 2014: 46, Winter 2015: 61

Error bars indicate standard error.

At the shallow (5m) site, differences in maximum skeletal diameter were not statistically significant between multigenetic and monogenetic clusters. However, survival and percent live tissue were both significantly higher in multigenetic clusters compared to monogenetic clusters ($p < 0.01$).

At the deep (12m) site, average (\pm SE) maximum skeletal diameter of colonies contained in multigenetic (“Mix”) clusters increased from 23.68 ± 1.19 cm to 39.40 ± 2.45 cm (Table 11). In the same time period, percent survival fell to $22.73 \pm 6.39\%$ while percent live tissue coverage declined to just $1.61 \pm 0.60\%$ (Figures 69 & 70). Colonies in the monogenetic clusters (“Single”) increased in size from 26.83 ± 1.47 cm in 2013 to 37.40 ± 1.86 cm in 2015. By the end of the study, only $24.59 \pm 5.60\%$ of the colonies in these structures had living tissue with an average live tissue coverage of $5.26 \pm 1.67\%$.

Table 11: Maximum skeletal diameter separated by resampling effort and genetic composition for deep (12m) site clusters. Non-stacked structures only.

| Genetic Comp. | Sampling | # Days Deployed | Average Skeletal Maximum Diameter (cm) | Standard Error (cm) | Sample Size |
|------------------------|-------------|-----------------|--|---------------------|-------------|
| Multi-genetic Clusters | Winter 2013 | 71 | 23.68 | 1.19 | 66 |
| | Summer 2014 | 221 | 29.81 | 1.40 | 52 |
| | Autumn 2014 | 319 | 37.36 | 2.56 | 22 |
| | Winter 2015 | 439 | 39.40 | 2.45 | 10 |
| Mono-genetic Clusters | Winter 2013 | 71 | 26.83 | 1.47 | 76 |
| | Summer 2014 | 221 | 34.40 | 1.87 | 53 |
| | Autumn 2014 | 317 | 43.32 | 6.59 | 19 |
| | Winter 2015 | 441 | 37.40 | 1.86 | 15 |

For the deep (12m) site, differences in maximum skeletal diameter, survival, and percent live tissue were not statistically significant between multigenetic and monogenetic treatments.

INDIVIDUAL GENOTYPE ANALYSES FOR CLUSTERS AT THE SHALLOW (5m) SITE

Survival and percent tissue coverage were analyzed for individual genotypes within clusters at the shallow site to determine if the performance any one genotype or individual cluster was exerting disproportionate influence on the results described above. Colonies transplanted into multigenetic structures were combined and given a “Mix” genotype designation to determine average survival and percent live tissue coverage because sample sizes were small. Average (\pm SE) percent survival for “Mix” colonies decreased to $86.11 \pm 5.85\%$ by 2015 (Figure 71). Survivorship for single genotype clusters fell to $55.00 \pm 11.41\%$ for U21 corals, $36.36 \pm 15.21\%$ for U39, and $69.23 \pm 13.32\%$ for U69. Percent live tissue declined to $57.39 \pm 6.78\%$ for “Mix”, $28.00 \pm 6.94\%$ for U21, $5.00 \pm 3.57\%$ for U39, and $45.38 \pm 10.55\%$ for U69, respectively (Figure 72).

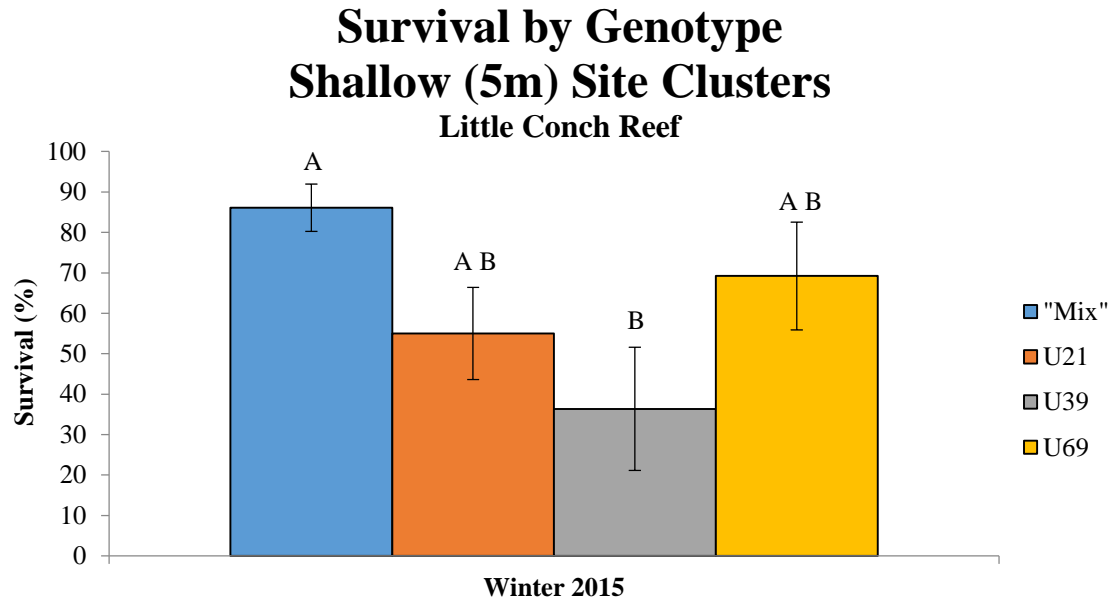


Figure 77: Winter 2015 percent survival separated by genotype for shallow (5m) clusters. Non-stacked structures only comparing “Mixed” and single genotype clusters. Percent survival = $((\# \text{ alive} / \# \text{ total}) * 100)$
 “Mix”: 36 colonies, U21: 20, U39: 11, U69: 13
 Error bars indicate standard error. Letters indicate statistical significance.

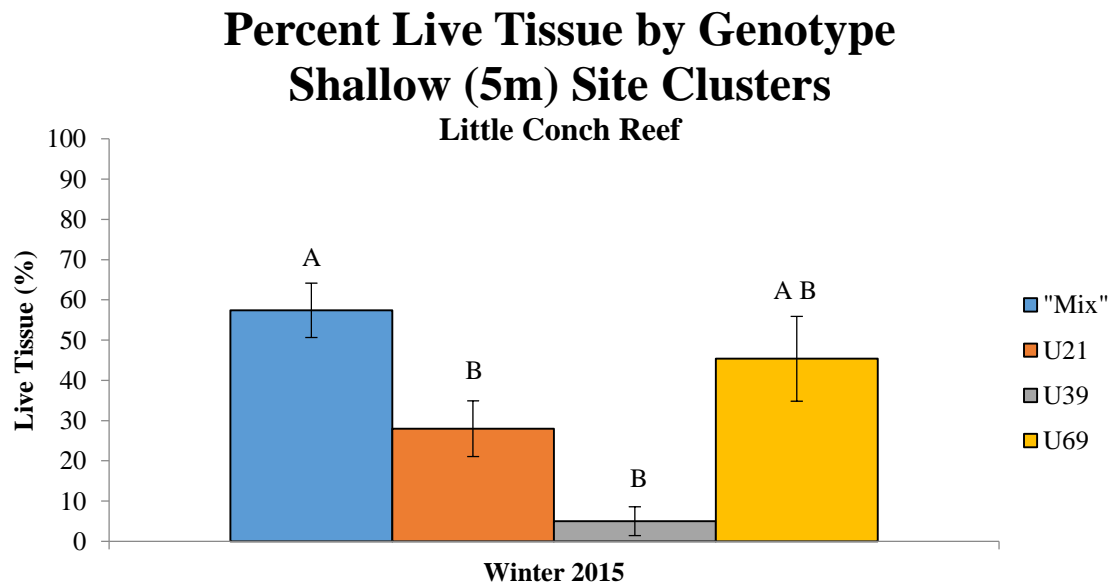


Figure 78: Winter 2015 percent live tissue separated by genotype for shallow (5m) clusters. Non-stacked structures only comparing “Mixed” and single genotype clusters. “Mix”: 36 colonies, U21: 20, U39: 11, U69: 13
 Error bars indicate standard error. Letters indicate statistical significance.

With respect to genotype at the shallow (5m) clusters, “Mix” colonies had a greater average survivorship by 2015 than U39 colonies ($p < 0.01$). “Mix” colonies also had more live tissue, on average, than U21 and U39 colonies ($p < 0.05$ and $p < 0.01$, respectively).

With respect to individual clusters (Figure 7), mean (\pm SE) percent survival of LCS_C14 ranked the lowest at 0%, followed by LCS_C12 at $28.57 \pm 18.44\%$, and LCS_C13 at $50.00 \pm 28.87\%$ (Figure 73). The top three included LCS_C01 at 100%, LCS_C08 at 100%, and LCS_C03 at $92.31 \pm 7.69\%$. A similar ranking was observed for percent live tissue coverage (Figure 74). The bottom three are LCS_C14 (0%), LCS_C12 ($1.43 \pm 0.92\%$), and LCS_C13 ($11.25 \pm 9.66\%$), while the top three are LCS_C01 ($82.50 \pm 5.69\%$), LCS_C03 ($72.69 \pm 10.68\%$), LCS_C11 ($54.00 \pm 15.44\%$).

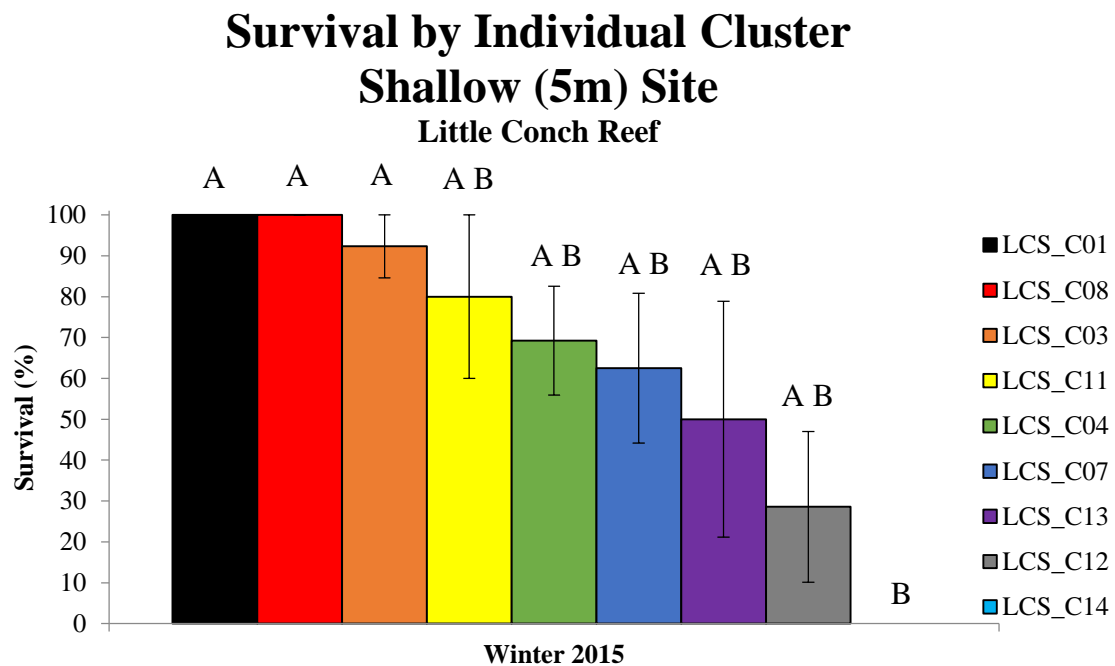


Figure 79: Winter 2015 percent survival separated by individual cluster for the shallow (5m) site.

Non-stacked structures only. Winter 2015 resampling only.

Percent survival = $((\# \text{ alive} / \# \text{ total}) * 100)$

LCS_C01: 10 colonies, C03: 13, C04: 13, C07: 8, C08: 11, C11: 5, C12: 7, C13: 4, C14: 9

Error bars indicate standard error. Letters indicate statistical significance.

Live Tissue by Individual Cluster Shallow (5m) Site Little Conch Reef

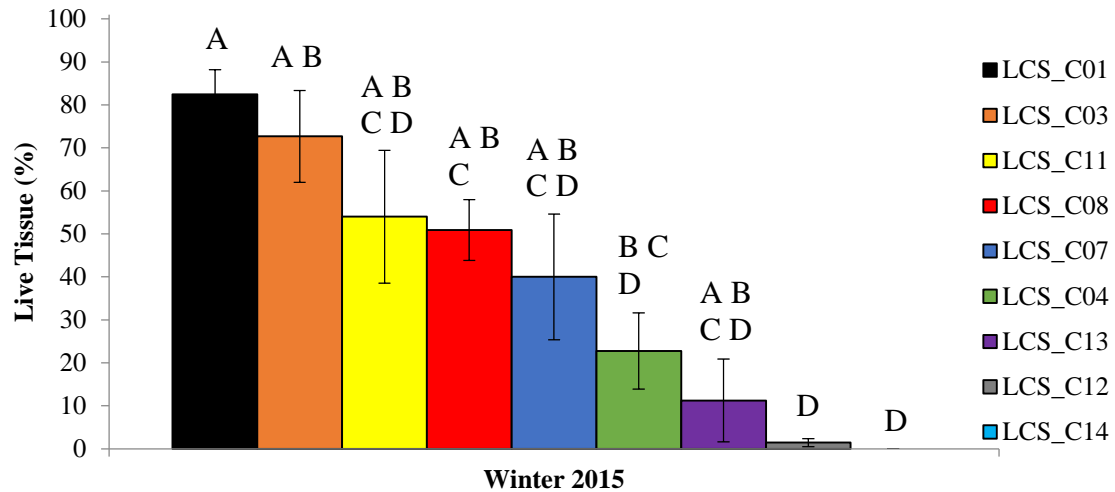


Figure 80: Winter 2015 percent live tissue separated by individual cluster for the shallow (5m) site.

Non-stacked structures only. Winter 2015 resampling only.

LCS_C01: 10 colonies, C03: 13, C04: 13, C07: 8, C08: 11, C11: 5, C12: 7, C13: 4, C14: 9

Error bars indicate standard error. Letters indicate statistical significance.

Colonies in LCS_C14 had a significantly lower average percent survival than those in LCS_C01 ($p < 0.001$), LCS_C03 ($p < 0.01$) and LCS_C08 ($p < 0.001$). Regarding percent live tissue coverage, LCS_C14 underperformed relative to LCS_C01, LCS_C03 and LCS_C08 ($p < 0.01$ for each). LCS_C12 has significantly less live tissue compared to the same three as LCS_C14 ($p < 0.05$ for each). LCS_C01 had more live tissue than LCS_C04 ($p < 0.05$). It is important to note that the highest performing clusters in both percent survival and live tissue coverage are located on the south side of the site, while the lowest performing are on the north side (Figure 7).

STACKING AT THE SHALLOW (5m) LITTLE CONCH REEF EXPERIMENT

On June 6, 2014, 963 *Acropora cervicornis* colonies were added to existing structures at the shallow (5m) site in an attempt to create larger structures. The idea tested was that larger structures might handle disturbances better than individual colonies, resulting in higher survival. Seven clusters and one thicket received additional colonies.

Since monitoring individual colonies in a stacked configuration was impossible due to difficulty identifying where each colony stopped, the structures were tracked as a unit.

After stacking, the average (\pm SE) maximum structure diameter for the clusters was 116.71 ± 9.15 cm. By the Winter 2015 sampling, the average maximum structure diameter was reduced to 100.29 ± 8.02 cm. Several pieces of the clusters were found lying near their parent structures with fragmented colonies still secured to each other by plastic zip-ties. Average live tissue coverage in the clusters was just $2.5 \pm 1.20\%$ by January 2015. The thicket underwent a similar reduction in maximum size and percent live tissue (400 cm to 360 cm, and 95% to 5%, respectively). Due to the small sample size, statistical comparisons were not possible.

2014 BLEACHING AT THE LITTLE CONCH REEF EXPERIMENT

During summer 2014, an intense bleaching event was observed across the Florida Keys (Coral Reef Watch 2015). Two metrics to assess bleaching were recorded: frequency (percentage of colonies demonstrating any level of bleaching) and severity (estimate of percent tissue per colony exhibiting bleaching). At the shallow (5m) site, $82.01 \pm 2.49\%$ of the colonies bleached with an average severity of $53.04 \pm 2.66\%$ (Table 12). Colonies at the deep (12m) site recorded a $62.38 \pm 4.84\%$ bleaching frequency with an average severity of $44.17 \pm 5.02\%$. Difference in bleaching frequency was significant ($p < 0.001$) while severity was not.

Table 12: Bleaching frequency and severity separated by depth. Non-stacked structures only.

| Site | Sampling | # Days Deployed | Bleaching Frequency (%) | Standard Error (%) | Bleaching Severity (%) | Standard Error (%) | Sample Size |
|--------------|-----------|-----------------|-------------------------|--------------------|------------------------|--------------------|-------------|
| Shallow (5m) | Fall 2014 | 334 | 82.01 | 2.49 | 53.04 | 2.66 | 239 |
| Deep (12m) | Fall 2014 | 316 | 62.38 | 4.84 | 44.17 | 5.02 | 101 |

Colonies contained in clusters at the shallow site recorded an average bleaching frequency of $54.55 \pm 6.18\%$ with an average severity of $19.32 \pm 4.37\%$ (Table 13). Thickets at this depth had an observed bleaching frequency of $92.49 \pm 2.01\%$ and a

severity of $61.93 \pm 2.78\%$. Both the difference in frequency and severity are significant (both $p < 0.0001$, respectively). At the deep site, $70.73 \pm 7.19\%$ of the colonies contained in the clusters bleached compared to 56.67% in the thickets. Average bleaching severity in the clusters was $54.21 \pm 6.94\%$ and $34.63 \pm 6.98\%$ in the thickets. Neither of these two metrics were statistically significant at the deep site, indicating that the clusters and thickets at this depth bleached similarly.

Table 13: Bleaching frequency and severity separated by depth and structure. Non-stacked structures only. Autumn 2014 sampling.

| Structure | # Days Deployed | Bleaching Frequency (%) | Standard Error (%) | Bleaching Severity (%) | Standard Error (%) | Sample Size |
|-----------------|-----------------|-------------------------|--------------------|------------------------|--------------------|-------------|
| Shallow Cluster | 334 | 54.55 | 6.18 | 19.32 | 4.37 | 66 |
| Shallow Thicket | 334 | 92.49 | 2.01 | 61.93 | 2.78 | 173 |
| Deep Cluster | 318 | 70.73 | 7.19 | 54.21 | 6.94 | 41 |
| Deep Thicket | 314 | 56.67 | 6.45 | 34.63 | 6.98 | 60 |

The multigenetic clusters at the shallow site bleached with both a greater frequency and severity compared to their monogenetic counterparts. $68.75 \pm 8.32\%$ of the colonies in the multigenetic structures bleached with an average severity of $22.22 \pm 6.03\%$, compared to $41.18 \pm 8.57\%$ of colonies in the monogenetic units, with an average severity of $17.31 \pm 6.17\%$ (Table 14). Only the difference in frequency was statistically significant for the shallow site ($p < 0.05$). At the deep site, monogenetic clusters fared worse. $78.95 \pm 9.61\%$ of colonies in the monogenetic structures bleached versus $63.64 \pm 10.50\%$ in the multigenetic clusters. Bleaching severity was also greater in the monogenetic units than their multigenetic counterparts ($61.39 \pm 8.85\%$ and $47.75 \pm 10.50\%$, respectively). However, neither the difference in bleaching frequency nor severity was statistically significant.

Table 14: Bleaching frequency and severity within clusters separated by depth and genetic composition. Non-stacked structures only. Autumn 2014 sampling.

| Genetic Compos. | # Days Deploy | Bleaching Frequency (%) | Standard Error (%) | Bleaching Severity (%) | Standard Error (%) | Sample Size |
|-------------------|---------------|-------------------------|--------------------|------------------------|--------------------|-------------|
| Shallow Multigen. | 334 | 68.75 | 8.32 | 22.22 | 6.03 | 32 |
| Shallow Monogen. | 334 | 41.18 | 8.57 | 17.31 | 6.17 | 34 |
| Deep Multigen. | 319 | 63.64 | 10.50 | 47.75 | 10.50 | 22 |
| Deep Monogen. | 317 | 78.95 | 9.61 | 61.39 | 8.85 | 19 |

To take a closer look at the genetic component of the bleaching frequency differences at the shallow site clusters, they were analyzed based on individual genotypes and by individual cluster. The U69 genotype displayed the greatest bleaching frequency ($75.00 \pm 13.06\%$, Table 15). None of the U39 colonies bleached, resulting in a statistically significant difference when compared to U69 or “Mix” (both $p < 0.01$, Figure 75). Multigenetic clusters LCS C01 and C03 and monogenetic cluster C11 located on the south side of the site recorded the highest bleaching frequencies ($83.33 \pm 11.24\%$, $92.31 \pm 7.69\%$, and 100% , respectively, while LCS C04 on the north end did not bleach at all (Table 16). It is important to note that though LCS C14 recorded no bleaching, only two living colonies remained at the time of the Autumn 2014 sampling. Also, despite ultimately recording the highest mean percent survival and live tissue coverage by the end of the experiment, the south end of the site had the highest rates of bleaching frequency.

Table 15. Bleaching frequency and severity separated by genotype at the shallow (5m) site clusters. Non-stacked structures only. Autumn 2014 sampling.

| Genotype | Bleaching Frequency (%) | Standard Error (%) | Sample Size | Bleaching Severity (%) | Standard Error (%) | Sample Size |
|----------|-------------------------|--------------------|-------------|------------------------|--------------------|-------------|
| Mix | 68.75 | 8.32 | 32 | 22.22 | 6.03 | 18 |
| U21 | 41.67 | 14.86 | 12 | 7.14 | 7.14 | 7 |
| U39 | 0.00 | 0.00 | 10 | 0.00 | 0.00 | 10 |
| U69 | 75.00 | 13.06 | 12 | 44.44 | 13.03 | 9 |

Bleaching Frequency by Genotype Shallow (5m) Site Clusters Little Conch Reef

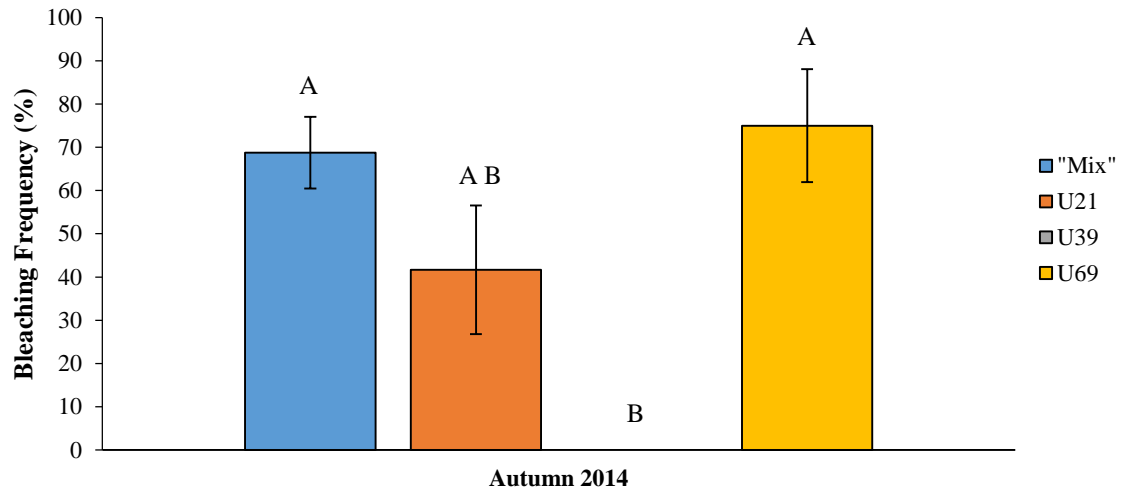


Figure 81. Autumn 2015 bleaching frequency separated by genotype at the shallow (5m) site clusters. Non-stacked structures only. Autumn 2014 sampling.

Bleaching frequency = ((# bleached/# total)*100)

"Mix" = 32 colonies, U21 = 12, U39 = 10, U69 = 12

Error bars indicate standard error. Letters denote statistical significance.

Table 16. Bleaching frequency and severity separated by individual cluster at the shallow (5m) site. Non-stacked structures only. Autumn 2014 sampling.

| Structural Identifier | Bleaching Frequency (%) | Standard Error (%) | Sample Size | Bleaching Severity (%) | Standard Error (%) | Sample Size |
|-----------------------|-------------------------|--------------------|-------------|------------------------|--------------------|-------------|
| LCS C01 | 83.33 | 11.24 | 12 | 33.33 | 10.54 | 6 |
| LCS C03 | 92.31 | 7.69 | 13 | 40.00 | 10.00 | 5 |
| LCS C04 | 0.00 | 0.00 | 7 | 0.00 | 0.00 | 7 |
| LCS C07 | 62.50 | 18.30 | 8 | 41.67 | 20.07 | 6 |
| LCS C08 | 50.00 | 16.67 | 10 | 10.00 | 10.00 | 5 |
| LCS C11 | 100 | 0.00 | 4 | 50.00 | 0.00 | 3 |
| LCS C13 | 0.00 | 0.00 | 10 | 0.00 | 0.00 | 10 |
| LCS C14 | 0.00 | 0.00 | 2 | 0.00 | 0.00 | 2 |

DISCUSSION

This project had two main objectives. First, to sample a subset of past Coral Restoration Foundation projects in the Upper Florida Keys using historical photographs and present-day *in situ* resampling to evaluate the number, sizes, and condition of transplanted colonies. This work provided a multi-year dataset for *Acropora cervicornis* coral colonies raised in an offshore nursery, then transplanted in multiple habitat types over a period of seven years. Variables included coral genotype, location, and habitat. Second, to monitor a coral transplant experiment at Little Conch Reef to evaluate the effects of depth, colony density, and genetic composition on growth, survivorship, and condition of nursery-raised *A. cervicornis*.

HISTORICAL PHOTOGRAPHIC ANALYSES

The photographic record provided by the Coral Restoration Foundation for this project covered 2,428 colonies from 17 projects over seven years, including six reefs, three habitat types, and 38 genotypes (Table 1). The projects began in 2007 and continued through 2013, extending from Dry Rocks in the north to Conch Reef in the south (Figure 82). The photographs generally followed each project for approximately two years. *In situ* resampling of 13 of the projects was conducted in 2014 using SCUBA. Generalizations from the photographic analyses include: 1) maximum size of *Acropora cervicornis* transplants on these reefs was approximately 40cm in diameter; 2) mortality increased after approximately two years; 3) despite high mortality, some colonies survived the duration of each project; and 4) frequent and long-term monitoring is required to assess factors that affect survival and condition.

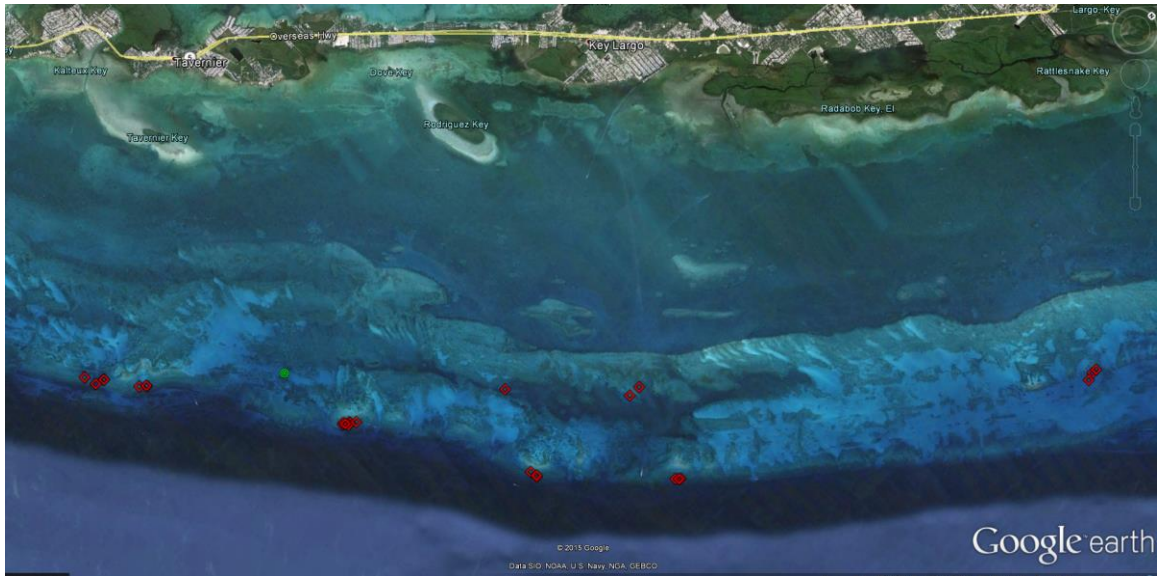


Figure 82. Location of 17 Coral Restoration Foundation projects utilized in photographic analyses shown as red diamonds. Green circle indicates Tavernier nursery.

The various Coral Restoration Foundation projects displayed a large range of maximum skeletal diameters but only a small percentage of projects (23.5%) had colonies that averaged larger than 40cm (Table 17). Three of the four projects with colonies that averaged larger than ~40cm (FR1, DR2, and FR2) were located on spur-and-groove habitats. The other (MD32) was on a hard-bottom site. All four sites had a maximum depth of 9m. Forty centimeters may represent a present-day functional maximum size, beyond which the colony may be more susceptible to fragmentation from wave action (Highsmith 1982). Another limitation may be a reduced rate of growth as the colony increases in size and metabolic resources are redistributed for other processes, such as reproduction (Lirman et al. 2014). Interestingly, 40cm maximum diameter is the largest size class in natural *Acropora cervicornis* populations in the Florida Keys (Miller et al. 2008).

Table 17. Summary of historical photographic analyses.

| Project ID | Habitat Type | Depth (m) | # of Transplants | Initial Trans. Date | Duration (yrs) | Survival (%) | Avg Max Diam (cm) |
|------------|---------------|-----------|------------------|---------------------|----------------|--------------|-------------------|
| ML12 | Spur & Groove | 7 | 18 | 2007-07-26 | 6.98 | 11.11 | 40.75 |
| PK1 | Spur & Groove | 5 | 18 | 2008-07-20 | 6.28 | 33.33 | 35.67 |
| WB1 | Patch Reef | 10 | 18 | 2008-08-27 | 1.57 | 11.11 | 30.73 |
| MD32 | Hard-Bottom | 9 | 18 | 2008-10-11 | 5.76 | 33.33 | 47.00 |
| DR1 | Spur & Groove | 9 | 18 | 2008-11-12 | 5.77 | 87.80 | 40.16 |
| MT1 | Patch Reef | 8 | 18 | 2009-01-22 | 1.07 | 5.56 | 20.81 |
| FR1 | Spur & Groove | 9 | 18 | 2009-04-02 | 5.42 | 77.78 | 67.71 |
| DR2 | Spur & Groove | 5 | 24 | 2009-07-13 | 5.22 | 66.67 | 56.38 |
| CS1 | Hard-Bottom | 8 | 24 | 2009-08-04 | 1.52 | 54.17 | 27.68 |
| CS2 | Hard-Bottom | 5 | 24 | 2009-10-16 | 1.32 | 87.50 | 23.61 |
| PK2 | Hard-Bottom | 9 | 24 | 2009-10-29 | 5.00 | 3.57 | 26.00 |
| FR2 | Spur & Groove | 9 | 24 | 2010-05-27 | 4.75 | 83.33 | 55.00 |
| MT2 | Patch Reef | 8 | 24 | 2010-07-30 | 0.95 | 58.33 | 23.36 |
| PKARRA | Hard-Bottom | 6 | 400 | 2012-04-24 | 2.56 | 72.00 | 28.94 |
| CNARRA | Hard-Bottom | 8 | 400 | 2012-05-12 | 2.67 | 22.95 | 25.97 |
| MLARRA | Spur & Groove | 7 | 400 | 2012-05-22 | 2.63 | 40.91 | 32.75 |
| DRARRA | Spur & Groove | 8 | 400 | 2012-06-05 | 2.63 | 46.28 | 20.99 |
| P2013 | Spur & Groove | 7 | 296 | 2013-07-19 | 1.32 | 69.78 | 30.35 |
| FR2013 | Spur & Groove | 10 | 161 | 2013-08-06 | 1.07 | 64.87 | 25.01 |
| CNC2013 | Hard-Bottom | 5 | 92 | 2013-12-17 | 1.10 | 81.93 | 29.12 |

The two to three year timeframe for maximum growth also corresponds with increased mortality in nearly all of the transplant projects. Survival and condition remained high until unknown events caused a significant decline (e.g. Figure 11). Unfortunately for many of the projects, large time gaps exist between the last recorded photograph and the *in situ* monitoring. Monitoring was insufficient during the projects to determine relationships with factors that cause mortality, such as bleaching (Eakin et al. 2010, Downs et al. 2013), disease (Aronson and Precht 2001), predation (Williams and Miller 2012), or thermal events (Lirman et al. 2011, Barton and Casey 2005). Despite these stresses, four of the ten projects with records longer than three years demonstrated greater than 66% survival at the 2014 sampling. All four of these projects were on spur-and-groove sites at either Dry Rocks or French Reef. Continued monitoring of the colonies transplanted in 2012 and 2013 is required to determine their mortality trajectory because they are currently within the two- to three-year window that marked the beginning of significant decline in the other projects.

Bruckner et al. (2008) demonstrated a similar increase in mortality after three years for rescued *Acropora palmata* fragments in Puerto Rico following the *Fortuna Reefer* grounding. In a transplant experiment conducted by Garrison and Ward (2008, 2012), mortality rate for *A. cervicornis* increased by approximately 150% each year the fragments were deployed, with a median survival of 2.4 years. During Coral Restoration Foundation maintenance visits, fragments (whether naturally separated from the parent colony or separated by a CRF diver) are reattached to the reef with epoxy to improve their chances of survival. These fragments were often not included in the photographic record. The inclusion of fragments for some of the projects (e.g. Dry Rocks #1, Figure 17) in the *in situ* monitoring in 2014 thus produced an over-estimation of natural survival. As asexual fragmentation represents a key reproductive strategy for staghorn coral, the approach employed by CRF makes good sense (Bowden-Kirby 2001, Edwards et al. 2010, Johnson et al. 2011) even if it complicates monitoring.

Anecdotal evidence suggests that fragments generated from transplanted colonies on the reef survive longer than their parent colonies (Ken Nedimyer, pers. comm.). Mortality of the parent colonies are affected by stresses on the reef that are not found in

the nursery, such as differences in depth, wave action, sedimentation, predation, current, or water temperature (Schopmeyer et al. 2012). Fragments are grown hanging from structures in the nursery for a year or more, so they are acclimated to mid-water conditions, compared to the benthos as transplants. Baums (2008) identified site adaptation from source populations as a key component in project planning to improve the chances of transplant survival. Colonies transplanted to locations similar to those from which they originated might survive better than those transplanted to different conditions. If a colony survives long enough on a reef in a new location, the colony may have time to acclimate to the new conditions, which could explain why fragments survive better than their parent colonies. Bliss (2015) demonstrated phenotypic plasticity over the course of 14 months in offshore-nursery-raised *Acropora cervicornis* colonies transplanted over large distances into environments different from which they were raised. This indicates that transplanted colonies are capable of acclimating to their new environment, which provides a survival advantage.

With the caveat that sample sizes were small in several projects, colonies transplanted to spur-and-groove habitats grew larger (e.g. Figure 49) and had higher percent survival and percent live tissue compared to those on hard-bottom or patch reef habitats (e.g. Figures 38 and 39) in four out of six years. The hydrodynamic conditions at the spur-and-groove sites may be more typical of the environments where *Acropora cervicornis* was historically found along the fore-reef (Goreau 1959, Bottjer 1980, Jaap 1984). In addition to the fore-reef, mid-channel patch reefs were also a habitat with extensive *A. cervicornis* (Jaap 1984) prior to White Band Disease and Bleaching, and it remains a habitat with abundant remaining populations today (Miller et al. 2008). However, due to the short-term nature of Coral Restoration Foundation monitoring and significant impacts from bleaching and thermal events, the results were not definitive regarding which habitat might be best suited for transplants.

Previous studies have demonstrated a relatively high degree of genetic diversity within *Acropora cervicornis* in Florida (Hemond and Vollmer 2010), and that there is a genetic component to disease resistance (Vollmer and Kline 2008), and site adaptation (Baums 2008, Bowden-Kirby 2001) in these corals. However, size and survivorship of

colonies in the photographic analyses based on genotype were not statistically significant in many of the projects. In the 2012 and 2013 projects, both years which included several hundred transplants, several genotypes appeared to have similar percent survival and percent live tissue. Colonies from U1, U51, U53, and U59 genotypes recorded greater than 75% survival in both years (Tables 3 and 5). U55 transplants ranked among the lowest performing colonies in both survival and percent live tissue. While clear differences in growth are observed in some genotypes in the nurseries, along with different responses to increased temperature that causes bleaching, the genetic component of survival and condition is not well understood. Small sample sizes due to difficulties identifying the genotype tags resulted in high variance, which made the genotype results suggestive, but not significant.

LITTLE CONCH REEF EXPERIMENT

Acropora cervicornis colonies were transplanted at two depths (5m and 12m) at Little Conch Reef between October and November 2013. The colonies were arranged in either clusters (14 colonies each, ten monogenetic and six multigenetic structures) or thickets (140 colonies each, ten colonies from each of the 14 genotypes arranged in monospecific subunits) (Figures 7 and 8). Additional colonies were stacked onto seven existing clusters and one thicket at the shallow site in June 2014 to create larger three-dimensional structures. The sites were monitored every three months through January 2015. The results suggest that: 1) maximum skeletal diameter was unaffected by any of the treatments; 2) percent survival and percent live tissue were higher at the shallow site compared to the deep site, and similarly, the clusters outperformed the thickets, and multigenetic clusters outperformed their monogenetic counterparts; 3) location within the shallow site had an impact on survival and condition, with clusters doing better on the south side than on the north; and 4) stacking did not positively impact growth, survival, or condition.



Figure 83. Location of Little Conch Reef experiment. Shallow (5m) is the left red diamond, while the deep (12m) site is the right red diamond. The Tavernier nursery is shown as a green circle.

Regardless of the experimental treatment (depth, transplant structure, or genetic composition), by the end of the study the transplanted colonies displayed no statistical difference in maximum skeletal diameter (Figure 64, Tables 7-11). Both the shallow and deep sites are within the historical and modern observed depth range for this species. High disease mortality at the deep site, along with the 2014 bleaching event and subsequent mortality, likely reduced growth of the colonies and clearly impacted survival and condition.

At the deep (12m) site, rapid disease-related mortality was observed at the start of the project. Within six months, almost half of the colonies had died (Figure 65). By the end of the study, few colonies remained alive, and those that were alive had little live tissue remaining (Figure 66). This was the first large transplant project conducted at the site. In the future “trial runs” or “pilot projects” should be considered to ensure site suitability before large-scale projects are undertaken. Histological analysis was conducted by Dr. Esther Peters (George Mason University). *Rickettsia*-like organisms were recorded in the mucocytes of polyps, along with full thickness tissue necrosis. Ciliates were often observed in the samples. In this case, the ciliates might be related to something in the sediment not found at the shallow site or could be a secondary infection (Sweet et al.

2014). Interestingly, colonies at the deep site bleached with less frequency and severity than their shallower counterparts (Table 12). Greater depth may have offered some protection from the warmer surface waters or UV penetration (Johnson et al. 2011). No temperature loggers or other physical oceanographic equipment were deployed, nor was the identity of any zooxanthellae clade determined as part of this study. Consequently, the cause of the bleaching differences remains unknown.

Colonies within clusters at the deep site had significantly greater percent survival and percent live tissue than colonies within thickets (Figures 67 and 68). With the presence of a pathogen at this depth, separation between the clusters may have assisted in slowing disease transmission, while high densities and large spatial coverage may have encouraged transmission in the thickets. A comparison could be drawn to the installation of “fire breaks” in forestry (Edwards et al. 2010). High densities of disease-susceptible colonies was a key contributing factor to the decline of the acroporids 40 years ago (Precht and Miller 2007, Aronson and Precht 2001, Williams and Miller 2012). At the shallow site, clusters offered no additional survival benefits but did offer a significant condition advantage following the autumn 2014 bleaching event (Figures 67 and 68). Stress from growing in close proximity to the other colonies may also be responsible for the greater bleaching frequency and severity within the thickets at this depth (Table 13). Johnson et al. (2011) proposed similar benefits of reduced disease transmission and predation pressure as a result of spacing in the design for coral nurseries.

At the deep site where a pathogen was the dominant stressor early in the project, a multigenetic cluster composition offered no advantage over a monogenetic one (Figures 69 and 70). At the shallow site where thermal stress was the dominant stressor, multigenetic clusters averaged both greater survival and percent live tissue coverage. Of the monogenetic clusters at the shallow site, those of U39 recorded the lowest survival and percent live tissue while U69 recorded the highest (Figures 71 and 72). However, both U39 clusters were located on the north end of the shallow site and both U69 clusters were at the south end. Thus, there could be a site factor involved, even though the distance between the two cluster types was 20m.

Multigenetic clusters may be more resilient to thermal stress. Resiliency is defined as the speed with which a system returns to a previous equilibrium following a disturbance (Pimm 1984). Multigenetic clusters at the shallow site bleached with a greater frequency than their monogenetic counterparts (Table 14) but also had the greatest percent survival and percent live tissue coverage (Figures 77 and 78). Reusch et al. (2005) demonstrated a similar increase in resilience in *Zostera marina* with increasing genetic diversity. Multigenetic plots of *Z. marina* had greater biomass, shoot density, and infaunal abundance than monogenetic plots after being subjected to heat stress. Reusch et al. (2005) proposed that genotypic diversity could replace species diversity in conferring resilience in species-depauperate systems. Within the monogenetic clusters, transplants from the U39 genotype had the lowest bleaching frequency while U69 had the greatest bleaching frequency (Figure 81). Intra-reef variability could be related site topography (Davis et al. 2011, Bowden-Kirby 2001, Bottjer 1980), but the mechanics relating bleaching to survival are not fully understood (Grottoli et al. 2014, Baker 2003).

Stacking colonies to create increased three-dimensional structure offered no benefit with respect to growth, survival or condition. The structures were complicated and time-consuming to erect and after construction, they collapsed under their own weight. Fragments containing zip-tied colonies were commonly seen around several shallow clusters. Due to logistical issues related to their construction combined with the lack of any measured benefit, this type of stacking does not appear to enhance growth or survival.

CONCLUSIONS

The objectives of the coral transplant projects and Little Conch Reef experiment were to enhance the natural recovery of the *Acropora cervicornis* populations and to learn about factors influencing the growth and survival of transplanted corals. The methods used are based on the premise that by artificially increasing the population sizes, nature can take over when conditions are right for recovery (van Oppen et al. 2015). However, a frequent challenge to coral restoration (Buddemeier et al. 2001, Graham et al. 2014) is that since all of the stressors and disturbances that have led to coral reef declines

over the last few decades remain, why should corals continue to be transplanted onto reefs where they were once found, but do not currently exist?

On the scale of months to a few years, restoration activities improve the aesthetic and ecological functions of coral reefs. A visually-pleasing reef attracts more recreational divers (Quinn and Kojis 2006, Jaap 1984). As the reef structure declines, so does its function as a feeding and breeding grounds for fish and other reef organisms (Alvarez-Filip et al. 2009, Gratwicke and Speight 2005). On the scale of decades, restoration activities increase the potential for natural recovery if or when conditions become appropriate. Despite the Florida Keys location at the northern limit the geographic distribution of *Acropora cervicornis* (Acropora BRT 2005), local management changes such as the installation of mooring buoys and navigational aids, updating of waste treatment infrastructure, and the use of no-take and other fisheries management strategies have reduced stress on the offshore reefs (National Marine Sanctuary Program 2007, Keller et al. 2009).

These local changes will not solve the problem of coral reef decline, but they can buy additional time until regional and global stressors can be addressed. Despite high mortality observed in the restoration projects, where many started with only 18 or 24 colonies, a few colonies survived to 2014. These surviving corals could provide a “seed population” for producing sexually dispersed larvae that are better adapted to survive mortality events (Johnson et al. 2011, Hemond and Vollmer 2010, Vollmer and Kline 2008) or asexual fragments that are better acclimated to the stressors found locally (Baums 2008, Bowden-Kirby 2008). Predictions of increased warming (IPCC 2014) that will likely result in the increased frequency and intensity of coral bleaching and disease virulence (Donner et al. 2005, Maynard et al. 2015) remain a major concern. Given the fast growth rate of *Acropora cervicornis* and the potential for suitable environmental conditions to arise, recovery in this species could be rapid (Precht and Miller 2007, Shinn et al. 2003). Evidence of persistence in this species (Miller et al. 2008) and expansion northward in Florida (Vargas-Ángel et al. 2003, Precht and Aronson 2004) suggest that it is too early to consider coral reefs a lost cause, and that coral restoration holds promise for enhancing recovery of *A. cervicornis*.

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